Phospholipid-Nucleoside Conjugates. 3.[1] Syntheses and Preliminary Biological Evaluation of 1-β-D-Arabinofuranosylcytosine 5'-Monophosphate-L-1,2-Dipalmitin and Selected 1-β-D-Arabinofuranosylcytosine 5'-Diphosphate-L-1,2-Diacylglycerols

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Several new phospholipid-*ara*-C conjugates have been prepared and tested as prodrugs of the parent *ara*-C. The new derivatives include *ara*-CMP-L-dipalmitin, *ara*-CDP-L-distearin, *ara*-CDP-L-dimyristin, *ara*-CDP-L-diolein, and the radioactively labeled derivative *ara*-CDP-L-di[1-^{14}C]palmitin. In addition, the unusually stable *ara*-CMP-L-dipalmitin-*N*-phosphoryldicyclohexylurea adduct was isolated as a crystalline solid (two diastereomers) in the reaction sequence to prepare *ara*-CMP-L-dipalmitin. The new prodrugs were solubilized by sonication methods and tested for their antiproliferative activity in vitro against mouse myeloma MPC-11 cells and against L1210 lymphoid leukemia. Such studies demonstrated that the antiproliferative activities of the prodrugs (as determined by ED_{50}) were less that *ara*-C on a molar basis. In the mouse myeloma cell line some evidence was obtained that the antiproliferative activity was related to the chain length of the fatty acid side chains in the prodrugs. In in vivo studies against L1210 lymphoid leukemia in mice, the prodrugs were shown to be much more effective than *ara*-C, with the overall efficacy apparently being independent of the length of the fatty acid side chain. Some evidence was obtained in the in vivo studies that the *ara*-CDP-L-dimyristin, which bears the shortest fatty acid side chain, was more toxic at the higher dosages than the longer chain length derivatives.

The nucleoside analogue *ara*-C[3] has proven useful as a chemotherapeutic agent for the clinical treatment of various types of cancer.[4-8] However, there are several problems associated with its use as a chemotherapeutic agent. Such problems include rapid catabolism to the biologically ineffective *ara*-U,[9] the eventual development by cells of resistance to the drug (due—at least in experimental tumor systems—to the inability of the cells to metabolize the *ara*-C to the *ara*-CTP, the actual cytotoxic agent which is a potent inhibitor of DNA polymerase[10,11]), and the excessive toxicity of *ara*-C due to activity against rapidly dividing normal cell types[4,12] (e.g., bone marrow).

Several approaches have been attempted to overcome these difficulties, including the judicious choice of a careful dosage regimen in order to lower toxic side effects.[13] After some initial attempts at improving the efficacy of *ara*-C by using a prodrug approach,[14-17] recent work in test systems has shown good success with prodrugs in which *ara*-C is covalently attached to either steroids[18-21] or phospholipids.[22-26] The phospholipid-nucleoside conjugates investigated to date (see Chart I) include *ara*-CDP-L-dipalmitin[22,23] (**1a**) [the 2'-epimer of the naturally occurring *ribo*-CDP-L-dipalmitin (**1b**)], *ara*-ADP-L-dipalmitin[23] (**2**), TuDP-L-dipalmitin[23] (**3**), *ara*-CDP-L-diacylglycerol (**1f**), in which the diacyl species is a mixed multispecies representing the fatty acid acyl chains found in egg lecithin,[25,26] and the diastereomeric mixture of *ara*-CDP-DL-dipalmitin[24-26] (**4a**), as well as the ether-linked diastereomeric derivatives *ara*-CDP-DL-bis(hexadecyloxy) propyl ether[25,26] (**4b**) and *ara*-CDP-DL-O^1-(octadecyloxy)-O^2-(octadecadienyloxy) propyl ether[25,26] (**4c**). In order to further investigate this extremely promising class of prodrugs, the study described herein was undertaken, with the aims of evaluating the effect of changing the nature of the phospholipid-drug linkage and to investigate the effect of specific changes in the fatty acid side chains on the pyrophosphate-linked derivatives. Accordingly, the target compounds were *ara*-CMP-L-dipalmitin (**5**), *ara*-CDP-L-distearin (**1c**) *ara*-CDP-L-dimyristin (**1d**), and *ara*-CDP-L-diolein (**1e**).

Chemistry. Several methods are potentially available for the preparation of nucleoside 5'-diphosphate derivatives

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(3) Abbreviations used are: *ara*-C, 1-β-D-arabinofuranosylcytosine; *ara*-U, 1-β-D-arabinofuranosyluracil; *ara*-CTP, 1-β-D-arabinofuranosylcytosine 5'-triphosphate; *ara*-CDP-L(or DL)-dipalmitin, 1-β-D-arabinofuranosylcytosine 5'-diphosphate-L-(or DL)-1,2-dipalmitin; *ribo*-CDP-L-dipalmitin, 1-β-D-ribofuranosylcytosine 5'-diphosphate-L-1,2-dipalmitin; *ara*-ADP-L-dipalmitin, 9-β-D-arabinofuranosyladenine 5'-diphosphate-L-1,2-dipalmitin; TuDP-L-dipalmitin, tubercidin 5'-diphosphate-L-1,2-dipalmitin; *ara*-CDP-DL-bis(hexadecyloxy) propyl ether, 1-β-D-arabinofuranosylcytosine 5'-diphosphate-DL-1,2-bis(hexadecyloxy) propyl ether; *ara*-CDP-DL-O^1-(octadecyloxy)-O^2-(octadecadienyl) propyl ether, 1-β-D-arabinofuranosylcytosine 5'-diphosphate-DL-O^1-(octadecyloxy)-O^2-(octadecadienyloxy) propyl ether; TBDMS, *tert*-butyldimethylsilyl; Lv, levulinyl; DCC, dicyclohexylcarbodiimide; TBAF, tetrabutylammonium fluoride; ED_{50}, dose required to effect 50% inhibition of growth, relative to a control; LD_{50}, dose required to effect 50% lethality, relative to a control.
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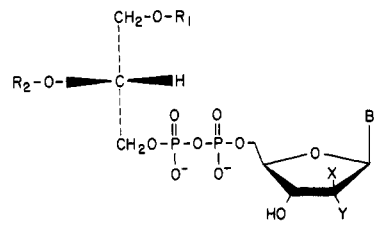
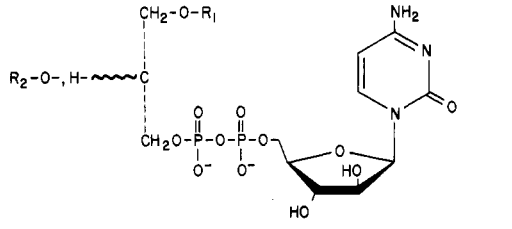
Chart I. Structures of Nucleoside-Phospholipid Drug Conjugates



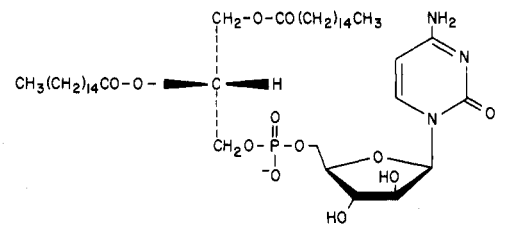
1 B = cytosine
(a) X = OH, Y = H, $R_1 = R_2 = CO(CH_2)_{14}CH_3$
(b) X = H, Y = OH, $R_1 = R_2 = CO(CH_2)_{14}CH_3$
(c) X = OH, Y = H, $R_1 = R_2 = CO(CH_2)_{16}CH_3$
(d) X = OH, Y = H, $R_1 = R_2 = CO(CH_2)_{12}CH_3$
(e) X = OH, Y = H, $R_1 = R_2 = CO(CH_2)_7-CH=CH-(CH_2)_7CH_3$
(f) X = OH, Y = H, R_1 = Predominantly multispecies saturated and monosaturated long fatty acyl chains; R_2 = Predominantly multispecies mono-, di-, and higher unsaturated long fatty acyl chains

2 B = adenine
X = OH, Y = H, $R_1 = R_2 = CO(CH_2)_{14}CH_3$

3 B = 7-deazaadenine (4-amino-7H-pyrrolo [2,3-d] pyrimidine)
X = H, Y = OH, $R_1 = R_2 = CO(CH_2)_{14}CH_3$



4 (a) $R_1 = R_2 = CO(CH_2)_{14}CH_3$
(b) $R_1 = R_2 = CH_2(CH_2)_{14}CH_3$
(c) $R_1 = CH_2(CH_2)_{16}CH_3$, $R_2 = CH_2(CH_2)_7(CH=CH-CH_2)_2(CH_2)_3CH_3$



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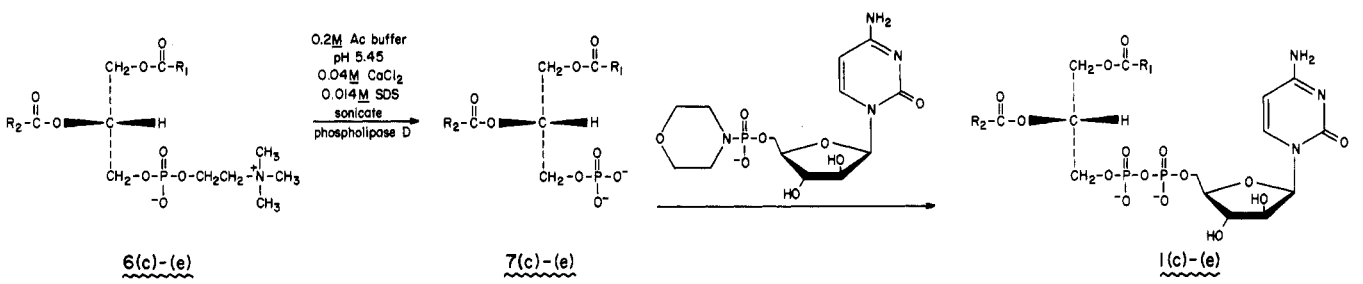
Scheme I. Preparation of Selected *ara*-CDP-L-1,2-Diacylglycerols



6(c)-(e) 7(c)-(e) 1(c)-(e)

(c) $R_1 = R_2 = -(CH_2)_{16}CH_3$
(d) $R_1 = R_2 = -(CH_2)_{12}CH_3$
(e) $R_1 = R_2 = -(CH_2)_7CH=CH(CH_2)_7CH_3$

such as **1–4**.[22,25,27-29] The more recent methods of choice utilize the activation of a nucleoside 5′-monophosphate by preparation of a nucleoside 5′-monophosphomorpholidate, which is then displaced by the phosphate moiety of the appropriate L-α-phosphatidic acid.[22,25,29] However, a major problem has been the availability of pure L-α-phosphatidic acids, which can be prepared chemically only via a synthetic scheme of several steps.[25,30-35] Dawson and Hemington[35] have described an enzymatic preparation of phosphatidic acid (having a mixture of egg lecithin derived fatty acid side chains) from egg lecithin using phospholipase D. More recently, Turcotte et al.[25] have presented a scale-up of the same reaction. The work described herein utilizes phospholipase D to prepare L-α-distearoylphosphatidic acid (**7c**), L-α-dimyristoylphosphatidic acid (**7d**), L-α-dioleoylphosphatidic acid (**7e**), and L-α-di[1-^{14}C]palmitoylphosphatidic acid from their respective L-α-phosphatidylcholine derivatives (**6c–e**) and their direct use in the synthesis of the respective *ara*-CDP-L-diacylglycerol derivatives (see Scheme I). Compounds **1c–e** were prepared in this fashion in overall yields of 40–50%. In addition, **1a** has been prepared on a larger scale than that described previously[22,25] using a similar enzymatic preparation of L-α-dipalmitoylphosphatidic acid.

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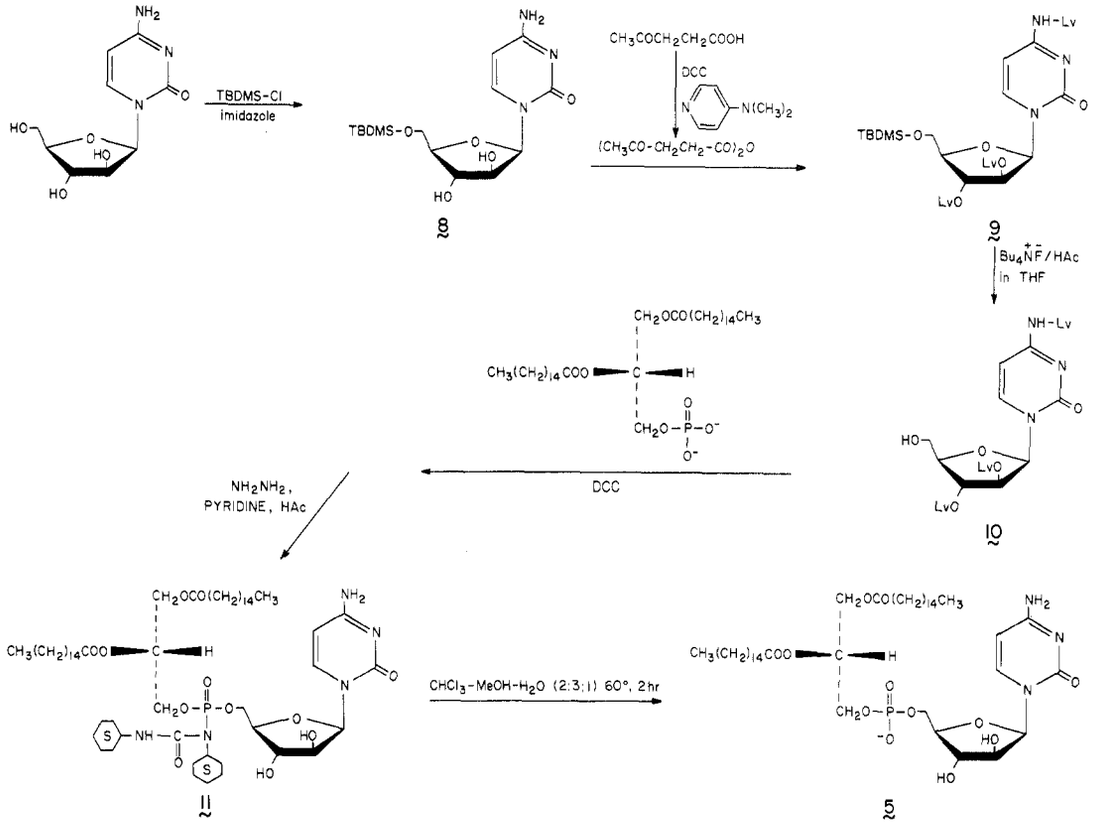
Scheme II. Preparation of *ara*-CMP-L-1,2-Dipalmitin



Preparation of the *ara*-CMP-L-dipalmitin derivative (**5**) presented a somewhat more difficult problem. Two synthetic schemes could be envisioned: one in which the final condensation was between 1,2-dipalmitin and a suitably protected (at the 2′,3′ and N^4 positions) *ara*-CMP, and another in which the final condensation was between L-α-dipalmitoylphosphatidic acid and a suitably protected (at the 2′,3′ and N^4 positions) *ara*-C. We chose the latter scheme because acyl migration in diacylglycerols is a well-documented phenomenon[36] and we wished to preclude contamination of the product (**5**) with undefined isomers. The choice of Scheme II was largely predicated on the selection of blocking groups for the hydroxyl groups at the 2′ and 3′ positions and for the exocyclic amine at N^4 of the *ara*-C moiety. Such blocking groups had to be removable under nonbasic conditions (otherwise concomitant saponification of the fatty acyl groups in the phospholipid moiety would be expected). Initial attempts at the use of TBDMS (removable using F^-[37]) or tetrahydropyranyl (removable under acidic conditions[38]) at the 2′ and 3′ positions in conjunction with either benzoyl (removable with hydrazine[39]) or dimethylaminomethylene (removable with dilute acid[40]) on the N^4 position were ultimately rejected due to extensive degradation of the product during the F^- or acid treatments necessary for the deblocking of the sugar protecting groups. The final blocking group of choice for the 2′, 3′ and N^4 positions was the levulinyl group. Thus, selective protection of the 5′-OH of *ara*-C was readily accomplished with *tert*-butyldimethylsilyl chloride in imidazole/DMF using procedures somewhat similar to those described earlier for *ara*-A.[41] The resulting 5′-*O*-TBDMS-*ara*-C (**8**) was acylated with levulinic anhydride [generated in situ from levulinic acid with DCC in the presence of 4-(dimethylamino)pyridine[42]] to produce the fully blocked 5′-*O*-TBDMS-2′,3′,N^4-triLv-*ara*-C (**9**). Selective removal of the 5′-TBDMS group was readily achieved with TBAF–acetic acid in tetrahydrofuran. In parallel with the observations of other workers,[43] we also noted the acyl migration (2′ → 5′) of the levulinyl group in these arabino derivatives when acetic acid was omitted during the TBAF treatment. The 2′,3′,N^4-trilevulinyl-*ara*-C (**10**) so obtained was then condensed with L-α-dipalmitoylphosphatidic acid (pyridinium salt) with DCC as condensing agent. The fully blocked product so obtained was not isolated but was treated with pyridine–H_2O and then hydrazine in pyridine-acetic acid to produce a highly nonpolar, uncharged compound, which was isolated pure by column chromatography on silica gel. This product was finally identified as the diastereomeric *N*-phosphoryldicyclohexylurea adduct (**11**). The structure was verified by elemental analysis, ^{31}P NMR (which showed two lines of equal intensity—corresponding to the two diastereomers—at 5.03 and 4.52 ppm downfield of the H_3PO_4 reference[44]), ^1H NMR (which showed a doubling

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Table I. Physical Data

no.	NMR data[a]	UV,[b] nm (ε)	formula	anal.[c]
1c	($CDCl_3$-CD_3OD-D_2O, 2:3:1) 8.03 (d, H6, J_{6-5} = 7.12 Hz), 6.18 (d, H1′, $J_{1'-2'}$ = 5.08 Hz), 6.10 (d, H5, J_{5-6} = 7.12 Hz), 5.28 (m, glycerol CH), 4.82-3.95 (m, HDO + H2′, H3′, H4′), 1.29 (m's, stearoyl CH_2's), 0.88 ("t", stearoyl CH_3's)	(i) 284 (14 600) (ii) 276 (10 390) (iii) 274 (9850)	$C_{48}H_{87}N_3O_{15}P_2Na_2 \cdot 6H_2O$	C, H, N, P
1d	($CDCl_3$-CH_3OD-D_2O, 2:3:1) 7.96 (d, H6, J_{6-5} = 6.78 Hz), 6.17 (d, H1′, $J_{1'-2'}$ = 5.09 Hz), 6.14 (d, H5, J_{5-6} = 6.78 Hz), 5.27 (m, glycerol CH), 4.75-3.93 (m, HDO + H2′, H3′, H4′, H5′, H5″ + glycerol CH_2's), 2.32 ("q", CH_2CO), 1.59 and 1.29 (m's, myristoyl CH_2's), 0.88 ("m", myristoyl CH_3's)	(i) 284 (13 020) (ii) 277 (9580) (iii) 274 (9170)	$C_{40}H_{71}N_3O_{15}P_2Na_2 \cdot 2H_2O$	C, H, N, P
1e	($CDCl_3$-CH_3OD-D_2O, 2:3:1) 8.03 (d, H6, J_{6-5} = 6.84 Hz), 6.18 (d, H1′ $J_{1'-2'}$ = 5.13 Hz), 6.09 (d, H5 J_{5-6} = 6.84 Hz), 5.301 (m, CH=CH + glycerol CH), 4.75-3.93 (m, HDO + H2′, H3′, H4′, H5′, H5″ + glycerol CH_2's), 2.33 ("q", CH_2CO), 2.01, 1.60, and 1.30 (m's, oleoyl CH_2's), 0.89 ("t", oleoyl CH_3's)	(i) 283 (14 010) (ii) 275 (9700) (iii) 274 (9170)	$C_{48}H_{83}N_3O_{15}P_2Na_2 \cdot 3H_2O$	C, H, N, P
5	($CDCl_3$-CH_3OD-D_2O, 2:3:1) 8.25 (d, H6, J_{6-5} = 7.7 Hz), 6.20 (d, H1′, $J_{1'-2'}$ = 5.05 Hz), 6.19 (d, H5, J_{5-6} = 7.7 Hz), 5.25 (m, glycerol CH), 4.80-3.91 (m, HDO + H2′, H3′, H4′, H5′, H5″ + glycerol CH_2's), 2.34 ("q", CH_2CO), 1.59 and 1.28 (m's, palmitoyl CH_2), 0.89 ("t", palmitoyl CH_3's)	(i) 284 (16 740) (ii) 275 (10 990) (iii) 274 (11 040)	$C_{44}H_{79}N_3O_{12}P_2Na_2 \cdot 2H_2O$	C, N, P; H[d]
8	(Me_2SO-d_6 + D_2O) 7.56 (d, H6, J_{6-5} = 7.42), 6.06 (d, H1′, $J_{1'-2'}$ = 4.64), 5.67 (d, H5, J_{5-6} = 7.42), 4.05 (m, H2′), 3.86 (m, H3′), 3.82 (m, H4′), 3.63 (m, H5′, H5″), 0.89 [s, $(CH_3)_3C$], 0.07 [s, $(CH_3)_2Si$]	(iv) 273 (8880)	$C_{15}H_{27}N_3O_5Si \cdot H_2O$	C, H, N
9	($CDCl_3$) 9.55 (s, NH), 8.11 (d, H6, J_{6-5} = 7.5 Hz), 7.38 (d, H5, J_{5-6} = 7.5 Hz), 6.24 (d, H1′, $J_{1'-2'}$ = 4.71 Hz), 5.61 (m, H2′), 5.25 (m, H3′), 4.07 (m, H4′), 3.83 (m, H5′, H5″), 2.80-2.59 (m, $COCH_2CH_2CO$), 2.21, 2.18, and 2.11 (3 s, Lv CH_3's), 0.93 [s, $(CH_3)_3C$], 0.11 [s, $(CH_3)_2Si$]			
10	($CDCl_3$) 9.73 (s, NH), 8.16 (d, H6, J_{6-5} = 7.55 Hz), 6.31 (d, H1′, $J_{1'-2'}$ = 4.25 Hz), 5.55 (m, H2′), 5.22 (m, H3′), 4.14 (m, H4′), 3.97 (m, H5′, H5″), 3.22 (t, OH5′), 2.95-2.50 (m, $COCH_2CH_2CO$), 2.23 (s, CH_3), 2.20 (s, CH_3), 2.15 (s, CH_3)	(v) 213 (13 240) 245 (9820) 308 (12 190) (iv) 213 (15 900) 250 (15 560) 298 (7000)	$C_{24}H_{31}N_3O_{11}$	C, H, N
11	($CDCl_3$-CD_3OD-D_2O, 1:1:0.04) 7.78 and 7.69 (2 d, H6, J_{6-5} = 7.4 Hz), 6.26 (2 d, apparent "t", H1′), 5.88 (2 d, H5, J_{5-6} = 7.4 Hz). Remaining lines too complex to interpret due to doubling of all resonances. ^{31}P NMR (CD_3OD) showed two lines at 5.03 and 4.52 ppm downfield from H_3PO_4	(iv) 273 (9000) (vi) 284 (14 100)	$C_{37}H_{102}N_5O_{12}P \cdot H_2O$	C, H, N, P

[a] ^1H NMR taken at 220 or 150 MHz; all shifts measured in δ from internal TSP (for **1c-e**, **5**, and **11**) or from Me_4Si (for **8-10**). ^{31}P data taken at 24.3 MHz. [b] (i) $CHCl_3$-MeOH-0.1 N HCl (2:3:1), (ii) $CHCl_3$-MeOH-H_2O (2:3:1), (iii) $CHCl_3$-MeOH-0.1 N-NaOH (2:3:1), (iv) MeOH, (v) 0.1 N HCl in MeOH, (vi) 0.1 N HCl in 90% aqueous MeOH. [c] Analyses within ±0.4% of calculated values. [d] H: calcd, 8.93; found, 9.40.

of the readily identified downfield resonances), IR spectroscopy [which showed identifiable bands at 1753 and 1700 (C=O), 1255 (P=O), 1168 (P-N), and at 1045 and 1020 cm^{-1} (P-O-C)], and UV spectroscopy (which showed a typical cytidine absorption spectrum). Structures analogous to **11** have previously been postulated[44,45] as reactive intermediates in phosphate diester syntheses using DCC as condensing agent and have been observed spectroscopically (^{31}P NMR) in reaction mixtures.[44] However, their usually facile hydrolysis in pyridine-H_2O has prevented their isolation and characterization by analytical methods. The reason for the unusual stability to hydrolysis of **11** is unclear, but it may be related to the formation of aggregates in which penetration of H_2O is precluded for steric reasons. The hydrolysis of **11** to produce *ara*-CMP-L-dipalmitin (**5**) was closely examined, and several straightforward methods were found to be successful. Complete hydrolysis (as judged by TLC) could be effected by either (i) BuOH-acetic acid acid-H_2O (5:2:3) at room temperature for 3 days, (ii) 10 M H_2O in pyridine at room temperature for 4 days, (iii) $CHCl_3$-MeOH-H_2O (2:3:1) at room temperature for 5 days, or (iv) $CHCl_3$-MeOH-H_2O (2:3:1) at 60 °C for 2 h. Method (iv) was utilized for all preparative purposes and led to the isolation and characterization of *ara*-CMP-L-dipalmitin (**5**) in 89% yield from **11**.

Biological Activity. Due to the insolubility in aqueous systems of most of the target compounds prepared in this study (**1d** and **1e** were the only derivatives that readily solubilized at room temperature], dissolution was obtained by the sonication techniques described earlier.[22,23] When these methods were used, *ara*-CDP-L-distearin (**1c**) remained slightly opalescent but was quantitatively (as estimated by UV) filtered through a 0.45-μm Millipore filter, and the filtered solution was used in in vitro studies as such. However, the *ara*-CMP-L-dipalmitin (**5**) resisted solubilization even when samples were mixed with several types of detergent prior to sonication. Finally, the best means of solubilization of **5** was achieved by mixing the sample with *ara*-CDP-L-dipalmitin prior to sonication. In this fashion, mixtures having ratios (by weight) of **5**:**1a** from 1:2 through 3:1 were used for the in vitro inhibition

(45) H. G. Khorana, "Some Recent Developments in the Chemistry of Phosphate Esters of Biological Importance", Wiley, New York, 1961.

Table II. In Vitro Antiproliferative Activity[a]

no.	compd	mouse myeloma cells[b] ED_{50}	mouse myeloma cells[b] LD_{50}	L1210 cells:[c] ED_{50}
	ara C	0.99 (0.85)	1.91	0.1
1a	*ara*-CDP-L-dipalmitin	65 (42.1)	68.5	9.6
1c	*ara*-CDP-L-distearin	195 (168.5)	164.5	20.0
1d	*ara*-CDP-L-dimyristin	15 (13.1)	15.2	16.0
1e	*ara*-CDP-L-diolein	62 (43.9)	54.8	27.0
5	*ara*-CMP-L-dipalmitin			13.4

[a] Values are in μM. [b] Values listed are for samples made up in H_2O; values in parentheses are for samples made up in 0.9% saline/0.1 mM Tris, pH 7.1. [c] Values are for samples made up in 0.5% Me_2SO in H_2O.

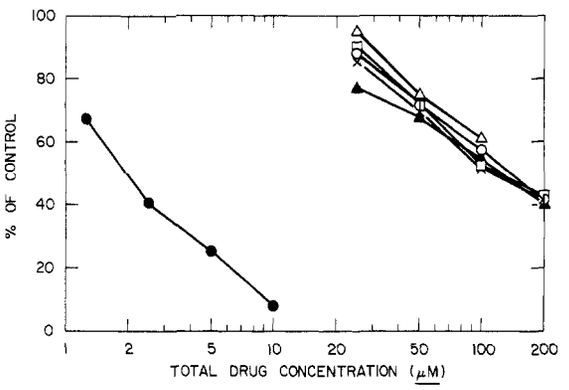

Figure 1. Inhibition of mouse myeloma MPC-11 cell line with various sonicated mixtures of *ara*-CMP-L-dipalmitin (**5**) and *ara*-CDP-L-dipalmitin (**1a**): (●) *ara*-C; (×) **1a**; (○) **5**:**1a** = 1:2; (△) **5**:**1a** = 1:1; (□) **5**:**1a** = 2:1; (▲) **5**:**1a** = 3:1.

studies against mouse myeloma MPC-11. However, for in vitro studies against L1210 lymphoid leukemia, the sample solutions were made up in 0.5% Me_2SO in H_2O with a more powerful sonicator. In this instance, *ara*-CMP-L-dipalmitin was effectively solubilized for in vitro testing.

In Vitro Studies. All the newly prepared derivatives were tested for antiproliferative activity against both mouse myeloma MPC-11 and L1210 lymphoid leukemia cell lines (see Table II). The use of these two cell lines provides for particular insight, since they differ in their sensitivity to the parent drug, *ara*-C.[22,23] In the mouse myeloma cell line, the testing of different ratios of **5**:**1a** did not markedly affect the overall ED_{50} of the samples, which were also similar to that obtained for **1a** alone (See Figure 1). This indicates extremely similar antiproliferative activities of **5** and **1a**. Because the latter is somewhat simpler to prepare, **5** was not investigated further in this cell line.

The ED_{50}'s of the diphosphate derivatives **1a**,**c**–**e** show interesting trends (see Table II). In the mouse myeloma cell line, the derivatives bearing fully saturated fatty acid side chains, namely **1a**,**c**,**d**, show a decrease in antiproliferative activity with increasing chain length; i.e., the order of antiproliferative activity follows *ara*-CDP-L-dimyristin (ED_{50} of 15 μM) > *ara*-CDP-L-dipalmitin (ED_{50} of 65 μM) > *ara*-CDP-L-distearin (ED_{50} of 195 μM). The *ara*-CDP-L-diolein derivative has an ED_{50} (62 μM) somewhat similar to that of the *ara*-CDP-L-dipalmitin derivative. The LD_{50}'s were also determined for **1a**,**c**–**e** and they show similar trends to the ED_{50} values (see Table II). Whether this order of biological effectiveness is due to different substrate affinities for the enzymes responsible for release of the parent drug *ara*-C (or the nucleotide *ara*-CMP[26]) from the prodrug,[46] to different rates of cellular uptake, or to other physicochemical factors is currently being investigated and will be published elsewhere.[47]

Because the samples for in vivo testing are made up in 0.9% saline and because the prodrugs **1a**,**c**–**e** show different aggregational characteristics when made up in buffered 0.9% saline,[48] it was of interest to examine samples made up in such a solution in the in vitro case. It is somewhat significant that the prodrugs so examined show very similar antiproliferative activities (as measured by ED_{50} values) as they do when made up in water (see Table II). It therefore appears that the different aggregational states of the prodrugs **1a**,**c**–**e**[48] are not responsible for the different cytotoxicities observed.

In the L1210 cell line, the effect of chain length on the antiproliferative activity was not as clear cut as in the mouse myeloma cell line; however, all the derivatives tested showed ED_{50} values somewhat higher (10–27 μM) than *ara*-C (0.1 μM). In the L1210 case, the order of antiproliferative activity was shown to be *ara*-CDP-L-dipalmitin (ED_{50} of 9.6 μM) > *ara*-CDP-L-dimyristin (ED_{50} of 16.0 μM) > *ara*-CDP-L-distearin (ED_{50} of 20.0 μM). The *ara*-CDP-L-diolein exhibited an ED_{50} of 27.0 μM, and in this series a value of 13.4 μM was obtained for *ara*-CMP-L-dipalmitin alone. It is of interest to note that the approximate equivalence of antiproliferative activity for **5** and **1a** that was suggested by the mixtures used against the mouse myeloma cell line (vide supra) was borne out in the L1210 case with separate samples.

It should be noted that *ara*-C and all its derivatives tested herein, with the exception of *ara*-CDP-L-dimyristin (which exhibited a similar activity in both cell lines) showed greater antiproliferative activity (on a molr basis) in the L1210 cell line than in the MPC-11 cell line. This is in accord with previous observations that have indicated the L1210 cell line to be more sensitive to *ara*-C than the mouse myeloma MPC-11 cell line.[22,23]

In Vivo Studies. Previous work from this laboratory[22,23] indicated that the efficacy of this type of prodrug was more apparent in in vivo studies than in in vitro. Accordingly, we have evaluated the derivatives described herein against L1210 lymphoid leukemia in mice. The effects of *ara*-C and its phospholipid conjugates against ip implanted L1210 lymphoid leukemia in $C_3D_2F_1/J$ mice are shown in Table III. The procedure was in accord with National Cancer Institute protocol,[49] with the exception that the inoculum in our studies contained 1×10^6 cells instead of 1×10^5 cells. Under our experimental conditions, the optimum dose found for *ara*-C was 20 (mg/kg)/day, and this dose given ip daily for 5 successive days showed an increase in life span (ILS) of 65%. However, treatments with the pyrophosphate-linked conjugate derivatives at the optimal dosages of 20–40 (mg/kg)/day for 5 days were clearly superior to *ara*-C with this particular dosage regimen. For example, *ara*-CDP-L-dipalmitin (**1a**), -distearin (**1c**), -dimyristin (**1d**), and -diolein (**1e**) at a dose of 40 (mg/kg)/day for 5 days exhibited ILS values of 188, 167, 153, and 153%, respectively. The monophosphate-linked conjugate (**5**) showed less activity, compared with

(46) See ref 24 for some of the enzymes of phospholipid metabolism for which *ara*-CDP-DL-dipalmitin is a substrate.
(47) M. MacCoss and T. Matsushita, manuscript in preparation.
(48) M. MacCoss, J. S. Edwards, T. M. Seed, and S. P. Spragg, manuscript in preparation.
(49) R. I. Geran, N. H. Greenberg, M. D. MacDonald, A. M. Schumacher, and B. J. Abbott, *Cancer Chemother. Rep., Part* 3, 3(2), 7 and 47 (1972).

Table III. Antitumor Activity in Mice Inoculated Intraperitoneally with L1210 Lymphoid Leukemia Cells[a]

no.	compd	dose,[b] [mg (μmol)/kg] / day × 5	wt change, g/mouse, on day 8	range	survival time, days, median (T/C)[c]	% ILS[d]	30-day survivors
	ara-C	10 (41.1)	+0.80	12-14	13.0/8.5	53	0
		20 (82.2)	+0.50	14-15	14.0/8.5	65	0
		40 (164.4)	+0.60	12-14	14.0/8.5	65	0
1a	*ara*-CDP-L-dipalmitin	25 (24.4)	+1.57	16-20	20.0/7.0	186	0
		40 (39.1)	−1.80	23 to >30	24.5/8.5	188	1
1c	*ara*-CDP-L-distearin	10 (9.5)	+1.20	15-17	15.0/7.5	100	0
		20 (19.0)	+1.10	15-21	18.0/7.5	140	0
		40 (37.9)	−0.40	16-28	20.0/7.5	167	0
1d	*ara*-CDP-L-dimyristin	10 (10.6)	+1.80	13-15	14.0/7.5	87	0
		20 (21.1)	+1.15	16-19	18.0/7.5	140	0
		40 (42.5)	+0.10	18-20	19.0/7.5	153	0
		60 (63.7)	−4.60	8-14	8.5/8.0	6	0
1e	*ara*-CDP-L-diolein	10 (9.5)	+1.50	13-15	14.0/7.5	87	0
		20 (19.1)	+1.40	15-18	16.0/7.5	113	0
		40 (38.1)	−0.10	18-21	19.0/7.5	153	0
		60 (57.1)	−3.85	15-20	17.0/8.0	113	0
5	*ara*-DMP-L-dipalmitin	50 (53.4)	+1.70	14-15	14.0/8.0	75	0

[a] $C_3D_2F_1$/J mice in groups of six (av wt 25 g) were inoculated ip with 1×10^6 cells of L1210 lymphoid leukemia. [b] Doses were started 24 h after tumor implantation and continued through day 5. [c] Median survival time of 24 control mice averaged 7.75 ± 0.65 (SD) days, and weight change on day 8 averaged +1.38 ± 0.95 (SD) g/mouse. [d] Percent ILS = (T/C − 1) × 100.

its corresponding diphosphate analogue (**1a**). The ILS value determined for the former (**5**) was 75% at a dose of 50 (mg/kg)/day for 5 days.

When the type of dosage regimen utilized in this study was used, toxicity (as reflected by weight loss and by a lowering of the ILS at higher dosages) was not observed in the *ara*-CDP–L-diacylglycerols described herein until a level of 60 (mg/kg)/day was reached. The most marked toxicity was with the *ara*-CDP–L-dimyristin derivative at such a dosage level. However, the toxicity of *ara*-C and of the conjugates at the optimal dosages [~40 (mg/kg)/day] was either minimal or not detectable with these dosage regimens. Thus, a much increased efficacy of these conjugates relative to the parent *ara*-C was apparent in vivo with the dosage regimen described above (Table III), although the antiproliferative activity measured in vitro showed the prodrugs to be less active. This trend has been observed previously[23,26] for this type of conjugate and it should be noted that some of the predicted advantages for these prodrugs (e.g., catabolic stability) would not be realized in the in vitro studies.[23,26]

In conclusion, the work described herein provides several new diastereomerically pure phospholipid derivatives of *ara*-C in which the fatty acid side chains have been varied and also in which the nature of the phospholipid–nucleoside linkage has been altered. The preliminary biological evaluation of these derivatives indicates that the antiproliferative activity in vivo against mouse myeloma MPC-11 is related to the nature of the side chain. In in vivo studies, each of the *ara*-CDP-L-diacylglycerols had a greater efficacy than the parent *ara*-C with the particular dosage regimens described, and some evidence was obtained that the toxicity was greatest for the *ara*-CDP–L-dimyristin derivatives. Overall, the promising biological data described herein and elsewhere[22,23,26] indicates the need for further investigation of this type of prodrug.

Experimental Section

Melting points were determined on a Thomas-Hoover melting point apparatus and are uncorrected. ^1H NMR spectra were recorded in the FT mode on either a Varian HR-220 or a Nicolet NT-150 WB spectrometer, and shifts are measured from Me_4Si or TSP as internal reference. ^{31}P NMR spectra were recorded in the FT mode on a Nicolet TT-14 spectrometer system equipped with a Nicolet multiobserve nuclei accessory (MONA) with H_3PO_4 as reference. UV spectra were recorded on a Beckman Model 25 spectrophotometer, and elemental analyses were determined by Galbraith Laboratories, Knoxville, TN. Evaporations were effected at 40 °C or lower with Buchi rotating evaporation under aspirator or mechanical oil-pump vacuum. Thin-layer chromatography (TLC) was performed on Merck silica gel 60, F-254, or on Eastman Chromagram 13181 silica gel with the solvents indicated. UV-absorbing compounds were detected by visualization under a UV lamp (254 nm) and phosphorous-containing compounds were detected with a modified Dittmer-Lester spray.[50] Column chromatography was effected with Merck silica gel 60 (70–230 mesh) or with the ion-exchangers indicated. Dry pyridine was prepared by distillation from CaH_2, then from chlorosulfonic acid (**caution** upon addition of the acid), and finally from KOH—the final distillate being collected and stored over activated 4A molecular sieves. Dry ethyl acetate was prepared by distillation from P_2O_5 and storage over molecular sieves. L-α-Dipalmitoylphosphatidic acid, L-α-dipalmitoylphosphatidyl choline, L-α-dimyristoylphosphatidyl choline, L-α-distearoylphosphatidyl choline, L-α-dioleoylphosphatidyl choline, and phospholipase D (Type III, from peanut, 130 units/mg of solid) were purchased from Sigma. L-α-Di[1-^{14}C]palmitoylphosphatidyl choline was purchased from Amersham. Solubilization of phosphatidyl choline samples were carried out with a Biosonik III sonicator (micro probe) at maximum power, and sonications for preparation of samples for biological evaluation were carried out as described previously with either a Kontes microultrasonic cell disrupter or a Biosonik IV.[22,23]

In vitro antiproliferative studies against mouse myeloma MPC-11 and L1210 lymphoid leukemia cell lines and in vivo studies against ip implanted L1210 lymphoid leukemia (1×10^6 cells) in C3D2F_1J mice were carried out as described previously.[22,23] The results are shown in Tables II and III.

1-β-D-Arabinofuranosylcytosine 5′-Diphosphate–L-1,2-Dimyristin (1d). L-α-Dimyristoylphosphatidyl choline (**6d**; 0.500 g, 0.00072 mol) was suspended in 30 mL of 0.2 M NaOAc/HOAc buffer, pH 5.45, containing 0.04 M $CaCl_2$. This material was subjected to sonication (Biosonik III) for 30 min. To this mixture was added 0.014 M sodium dodecyl sulfate solution (15 mL) and Et_2O (10 mL), and the total was added to a solution of phospholipase D (100 mg) in 50 mL of the above-mentioned 0.2 M NaOAc/HOAc buffer containing 0.04 M $CaCl_2$, the reaction vessel being a screw-top plastic container. The mixture was incubated at 30 °C and monitored by TLC (Merck; with $CHCl_3$–MeOH–H_2O–HOAc, 25:15:4:2, as solvent). After 22 h and again after 96 h, additional 20-mg increments of phospholipase D were added. After 10 days, the reaction was acidified to pH 1–1.5 with concentrated HCl and extracted with 100 mL of $CHCl_3$–MeOH (1:1) and then with $CHCl_3$ (100 mL). The combined organic layers were

(50) E. K. Ryu and M. MacCoss, *J. Lipid Res.*, **20**, 561 (1979).

evaporated to dryness to yield 0.45 g of a white solid (TLC showed the major spot to be **7d** slightly contaminated with trace amounts of **6d** and one other unidentified compound). This material was mixed with 1-β-D-arabinofuranosylcytosine 5′-monophosphomorpholidate, 4-morpholino-*N*,*N*′-dicyclohexylcarboxamidinium salt[22] (0.48 g, 0.0007 mol), and dissolved in dry pyridine (20 mL). The solution was evaporated to dryness in vacuo 5 times from dry pyridine and then was concentrated to ~15 mL from 50 mL of dry pyridine. This solution was stoppered and stored in a desiccator for 4 days with occasional shaking. The reaction mixture was then evaporated to dryness in vacuo, and the residue was coevaporated from toluene (4 × 20 mL). This residue was then dissolved in 30 mL of $CHCl_3$–MeOH–H_2O (2:3:1) and acidified to pH 3 with 0.1 N HCl. Two layers formed, and the aqueous layer was washed with $CHCl_3$ (2 × 15 mL). The combined organic layers were evaporated to dryness, and the residue was dissolved in $CHCl_3$–MeOH–H_2O (2:3:1) and applied to a DEAE-Sephadex (acetate form) column[51] (3.25 × 52 cm) packed in the same solvent. The column was eluted with 500 mL of $CHCl_3$–MeOH–H_2O (2:3:1) and then with a linear gradient (2 L in each reservoir) of 0–0.2 M ammonium acetate made up in the same solvent. Fractions containing the required product as judged by TLC (Merck; $CHCl_3$–MeOH–H_2O–HOAc, 25:15:4:2) were pooled and evaporated to ~120 mL. This mixture was extracted with $CHCl_3$ (500 mL), and the organic layer was evaporated to yield 0.325 g (50%) of **1d** as the diammonium salt. This material was dissolved in a minimum of $CHCl_3$–MeOH–H_2O (2:3:1) and converted to the disodium salt by passage down a column (2 × 12 cm) of Cellex-CM (Na^+ form) that had been packed in the same solvent mixture. The product was obtained as a tractable white precipitate by dissolution in $CHCl_3$ and addition of acetone. This was filtered off and washed well with acetone before being dried in vacuo over P_2O_5: mp, sample turns brown at 195–200 °C, with sharp melting at 209–210 °C.

1-β-D-Arabinofuranosylcytosine 5′-Diphosphate–L-1,2-Distearin (1c). This was prepared as described above for **1d**, starting from **6c**, to give **1c** in 40% yield: mp, sample slowly turns brown above 190 °C, with sharp melting at 204–205 °C.

1-β-D-Arabinofuranosylcytosine 5′-Diphosphate–L-1,2-Diolein (1e). This was prepared as described above for **1d**, starting from **6e** to give **1e** in 44% yield: mp, sample turns brown at 180–185 °C, with sharp melting at 204–205 °C.

1-β-D-Arabinofuranosylcytosine 5′-Diphosphate–L-1,2-Di[1-^{14}C]palmitin. This was prepared as described above for **1d**, starting from L-α-di[1-^{14}C]palmitoylphosphatidyl choline (50 μCi of 114 mCi/mmol) diluted with cold L-α-dipalmitoylphosphatidyl choline (0.043 g, 0.000057 mol). The overall yield was 30%, and the final product had a specific activity of 0.68 mCi/mmol.

5′-*O*-(*tert*-Butyldimethylsilyl)-1-β-D-arabinofuranosylcytosine (8). To a solution of *ara*-C (2.0 g, 0.0082 mol) and imidazole (1.237 g, 0.0181 mol) in dry DMF was added *tert*-butyldimethylsilyl chloride (1.363 g, 0.00904 mol), and the solution was stirred at room temperature in a desiccator overnight. The reaction mixture was evaporated to dryness in vacuo (one additional time from toluene), and water (150 mL) was added to the residue. After trituration, a white powder was formed, which was filtered off and washed with water to yield 4.8 g of crude product. This was recrystallized from ethyl acetate (~200 mL) containing a little MeOH (solution was cooled to 5 °C before filtering) to give 2.48 g (84.5%) of **8**. Product was homogeneous by TLC (Eastman) using the upper phase of ethyl acetate–1-propanol–H_2O (4:1:2) and had mp 230–231 °C.

5′-*O*-(*tert*-Butyldimethylsilyl)-2′,3′,*N*^{4}-trilevulinyl-1-β-D-arabinofuranosylcytosine (9). Levulinic acid (1.04 mL) was dissolved in dry ethyl acetate (25 mL), and DCC (2.1 g, 0.01 mol) was added. The reaction mixture was shaken occasionally while being stored in a desiccator at room temperature for 1.5 h. The mixture was then filtered directly onto a suspension of **8** (0.4 g, 0.00112 mol) and *N*,*N*-dimethylaminopyridine (0.16 g, 0.0013 mol) in dry ethyl acetate (25 mL), washing the filter pad (DCU) with more dry ethyl acetate (25 mL). This reaction was stirred at room temperature for 4.5 h when dry EtOH (4 mL) was added. After an additional 30 min, the precipitate (DCU) was filtered off, and the filtrate was extracted with ice-cold saturated $NaHCO_3$ solution (3 × 50 mL) and then water (3 × 50 mL) before being evaporated to dryness. The residue was dissolved in a minimum volume of $CHCl_3$ and applied to a column of silica gel (55 × 3 cm, wet packed in $CHCl_3$). The column was developed with $CHCl_3$ (1.2 L) and then with 5% MeOH in $CHCl_3$. Fractions containing the required product were combined and evaporated to dryness to give 0.713 g (97.8%) of **9**, which was homogeneous (R_f 0.63) by TLC (Merck) using EtOH–$CHCl_3$ (1:9) and by NMR, and was used directly for the preparation of **10**.

2′,3′,*N*^{4}-Trilevulinyl-1-β-D-arabinofuranosylcytosine (10). The foam of **9** prepared above (0.713 g, 0.0011 mol) was dissolved in a premade solution of 1 M TBAF in THF (5 ml) containing glacial acetic acid (0.5 mL). The reaction mixture was left at room temperature for 1.5 h and was then evaporated to dryness in vacuo (one additional time from toluene). TLC (Merck) using MeOH–$CHCl_3$ (1:9) indicated almost complete reaction, and the residue was dissolved in a minimum amount of $CHCl_3$ and applied to a silica gel column (43 × 3 cm), wet-packed in $CHCl_3$. The column was developed with $CHCl_3$ (300 mL), 5% MeOH in $CHCl_3$ (350 mL), and then 8% MeOH in $CHCl_3$ (400 mL). Fractions containing the required product were pooled and evaporated to dryness, yielding a clear gum. Crystallization from acetone–CCl_4 gave 0.433 g (74%) of analytically pure **10**, having mp 142–144 °C.

L-α-Dipalmitoylphosphatidic Acid Pyridinium Salt.[22] Commercially available L-α-dipalmitoylphosphatidic acid disodium salt (1.0 g, 0.00149 mol) was dissolved in 250 mL of $CHCl_3$–MeOH–pyridine–H_2O (3:3:1:1) and applied to a column of Dowex 50W × 8 (pyridinium) resin.[52] The required product was eluted with the solvent front, and the solution was evaporated to dryness [coevaporated several times from $CHCl_3$–MeOH (1:1)]. The residue was triturated with acetone, and the white solid was filtered off, washed with acetone, and dried over P_2O_5 in vacuo for 24 h.

1-β-D-Arabinofuranosylcytosine 5′-Monophosphate–L-1,2-Dipalmitin–*N*-Phosphoryldicyclohexylurea Adduct (11). A mixture of **10** (0.270 g, 0.0005 mol) and L-α-dipalmitoylphosphatidic acid pyridinium salt (0.356 g, 0.00051 mol) was dissolved in dry pyridine, and the mixture was rendered anhydrous by repeated evaporation from dry pyridine in vacuo. After addition of DCC (0.65 g, 0.00315 mol), the mixture was again evaporated to dryness in vacuo several times from dry pyridine. Finally, the volume was reduced to ~2 mL, and the mixture was left at room temperature in a desiccator for 4 days. Ice (~2 g) was then added, and the mixture was allowed to stand at room temperature overnight. The mixture was extracted with petroleum ether (30–60 °C; 3 × 5 mL) and then evaporated to dryness in vacuo. Pyridine was removed by several coevaporations in vacuo from toluene. The residue was dissolved in $CHCl_3$–MeOH (1:1, 5 mL) and stored at 4 °C overnight. The white precipitate of DCU so formed was filtered off and washed with the same solvent (2 × 2 mL). Combined filtrate and washings were evaporated to dryness, and the residue was dissolved in $CHCl_3$ and applied to a column (30 × 2.5 cm) of silica gel wet packed in $CHCl_3$. The column was developed with $CHCl_3$ and then 2% MeOH in $CHCl_3$, and fractions containing the major UV-absorbing product were pooled and evaporated to dryness. The residue was dissolved in 2 mL of pyridine–acetic acid (4:1), and hydrazine hydrate (0.1 mL) was added. After stirring for 2 h, TLC (Merck; $CHCl_3$–MeOH–H_2O, 65:25:3, as developer) showed that deblocking of the levulinyl groups was complete, and so the reaction mixture was evaporated to dryness. The residue was dissolved in $CHCl_3$ (30 mL) and applied to a column of silica gel, which was developed with 2% MeOH in $CHCl_3$ (500 mL) and then 10% MeOH in $CHCl_3$. Fractions containing the required product were pooled and evaporated to a small volume, and then MeOH was added. A white precipitate formed, which was filtered off and dried in

(51) The DEAE-Sephadex (acetate form) was washed with 50% aqueous MeOH and then MeOH prior to packing in $CHCl_3$–MeOH–H_2O (2:3:1).

(52) Prepared from Dowex 50WX8 (H^+) by washing with 50% aqueous pyridine, 50% aqueous MeOH, MeOH, and then $CHCl_3$–MeOH–pyridine–H_2O (3:3:1:1).

vacuo over P_2O_5. The yield was 0.120 g (22%), and the diastereomeric mixture of 11 so formed had mp 101–103 °C. TLC (Merck; $CHCl_3$–MeOH–H_2O, 65:25:3) showed one UV-absorbing spot, which was also positive using a phosphate spray reagent.[50] IR (KBr, disk) showed 1753, 1700 (C=O), 1255 (P=O), 1168 (P=N), and 1045, 1020 cm^{-1} (P–O–C) as identifiable bands.

1-β-D-Arabinofuranosylcytosine 5′-Monophosphate–L-1,2-Dipalmitin Sodium Salt (5). A solution of 11 (0.165 g, 0.00015 mol) in $CHCl_3$–MeOH–H_2O (2:3:1; 50 mL) was heated at 60 °C for 2 h. Monitoring by TLC (Merck, MeOH–$CHCl_3$, 3:7) indicated complete reaction, and the solution was evaporated to dryness. The residue was dissolved in $CHCl_3$–MeOH–H_2O (4:6:1; 100 mL) and applied to a column of DEAE-Sephadex (acetate form; 15 × 4 cm) packed in the same solvent.[53] Elution was initially with $CHCl_3$–MeOH–H_2O (4:1:1; 500 mL) and then with a linear gradient (700 mL in each reservoir) of 0–0.1 N ammonium acetate made up in $CHCl_3$–MeOH–H_2O (4:6:1). Fractions containing the required product were pooled and evaporated to small volume. Acetone (50 mL) was added, and the white precipitate of the NH_4^+ salt so obtained was filtered off andd then converted to the Na^+ salt by dissolution in $CHCl_3$–MeOH–H_2O (4:6:1) and passage down a Cellex-CM (Na^+ form) column 31 × 2.5 cm), packed, and developed in the same solvent. The fractions containing the required product were pooled and concentrated, and 5 was obtained as a white precipitate upon addition of acetone–H_2O (9:1). This material was filtered off, washed with acetone, and dried over P_2O_5 in vacuo for 20 h: yield 0.122 g (89%); mp 212–214 °C dec.

(53) The DEAE-Sephadex (acetate form) was prepared as described in ref 51, with the exception that the final equilibration and packing was carried out in $CHCl_3$–MeOH–H_2O (4:6:1).

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Synthesis and Antiviral Properties of Some 2′-Deoxy-5-(fluoroalkenyl)uridines

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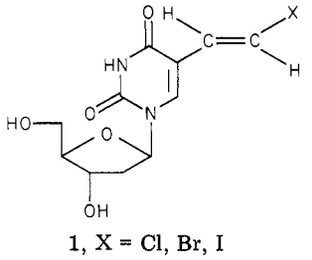
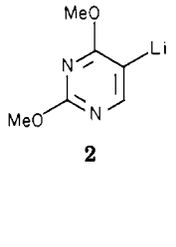
The following 5-substituted 2,4-dimethoxypyrimidines were synthesized: 5-(2,2,2-trichloro-1-hydroxyethyl), 5-(2,2,2-trichloro-1-fluoroethyl), 5-(2,2-dichloro-1-fluorovinyl) (**5**), and 5-(perfluoropropen-1-yl) (a mixture of *E* and *Z* isomers, **6** and **7**). Demethylation of **5** gave 5-(2,2-dichloro-1-fluorovinyl)uracil, and demethylation of the mixture of **6** and **7** gave some pure (*E*)-5-(perfluoropropen-1-yl)uracil. Compound **5** was converted into its 2′-deoxyribonucleoside (**12**) and its α-anomer by standard procedures. 2′-Deoxy-3,5-dilithio-3′,5′-*O*-bis(trimethylsilyl)uridine was reacted with the appropriate fluoroalkene to give the following 5-substituted 2′-deoxyuridines in low yield (6–24%): 5-(2-chloro-1,2-difluorovinyl) (a mixture of *E* and *Z* isomers, **15** and **16**, which were separated on a small scale), 5-(perfluoropropen-1-yl), 5-(perfluorocyclohexen-1-yl), and 5-(perfluorocyclopenten-1-yl). In these reactions, 2′-deoxy-5-(trimethylsilyl)uridine and 2′-deoxyuridine were also formed. The 5-substituted 2′-deoxyuridines were tested for activity against herpes simplex virus type 1. Compound **12** and the mixture of **15** and **16** had an ID_{50} of 20–26 μg/mL in Vero cells. The activity of the mixture resided in one isomer, which by analogy with the corresponding (*Z*)- and (*E*)-5-(2-bromovinyl)-2′-deoxyuridines was concluded to be the *Z* isomer (**16**).

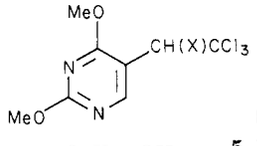
(*E*)-5-(2-Halogenovinyl)-2′-deoxyuridines (**1**) have been



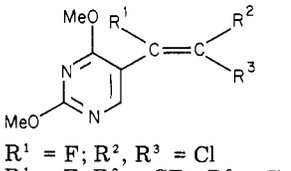
1, X = Cl, Br, I



2



3, X = OH
4, X = F



5, R^1 = F; R^2, R^3 = Cl
6, R^1 = F; R^2 = CF_3; R^3 = F
7, R^1 = F; R^2 = F; R^3 = CF_3

shown to be active against certain herpes viruses;[1] the bromo derivative is the most active, showing highly selective action against herpes simplex virus type 1 and against varicella zoster virus.[2] The compound has been extensively studied in vitro and in vivo and has shown promising results.[3] Similar compounds that also show high activity against herpes viruses are the (*E*)-5-(propen-1-yl)- and (*E*)-5-(3,3,3-trifluoropropen-1-yl) derivatives.[4] On the other hand, (*Z*)-5-(2-bromovinyl)-2′-deoxyuridine has only a very low activity, thus showing the critical nature of the configuration around the ethylenic double bond.[5] Obviously it is of interest to synthesize similar compounds and investigate their antiviral properties. In the present study, attention has turned to ob-

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