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Poly(vinyl alcohol) with Pending 5-Substituted Uracils

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5-Chloro-, bromo-, iodo-, *N*-ethylamino-, and *N,N'*-diethylamino-1-[(2'-dihydrogenphosphato)-ethyl]-uracil were synthesized from 1-(2'-hydroxyethyl)-uracil through the combination of halogenation, phosphorylation, and subsequent amination. These compounds were coupled with poly(vinyl alcohol) using *N,N'*-dicyclohexylcarbodiimide as a dehydrating agent to give polymers having 5-substituted uracils as pending groups.

In previous papers we have reported on the syntheses of oligonucleotide analogues from *N*-(2'-hydroxy-3'-dihydrogenphosphato)-propyl derivative of nucleic-acid bases.[1,2] These oligomers were very soluble in water and showed significant hypochromic effect, a typical spectroscopic behavior of natural polynucleotides. As another polymer we prepared poly(vinyl alcohol) having nucleic-acid bases as pending groups by the condensation of *N*-(2'-hydroxyethyl) derivatives of nucleic-acid bases with poly(vinyl al-

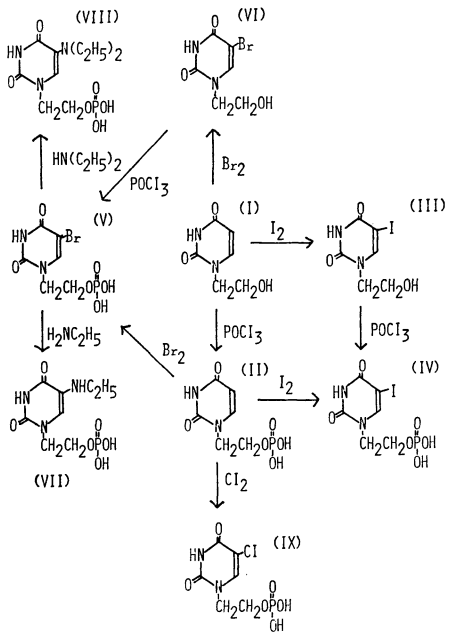
1) T. Seita, K. Yamauchi, M. Kinoshita, and M. Imoto, This Bulletin, **45**, 926 (1972).

2) T. Seita, K. Yamauchi, M. Kinoshita, and M. Imoto, *Makromol. Chem.*, **154**, 255 (1972).

cohol). The polymers obtained were also hygroscopic and very soluble in water and showed the same hypochromic effect.[3] The excellent solubility makes it possible to use these polymers for biological systems as polymeric drugs. A most interesting utilization might be found in the incorporation of 5-halo-uracils, especially the important anti-cancer material 5-fluoro-uracil, into polymers.

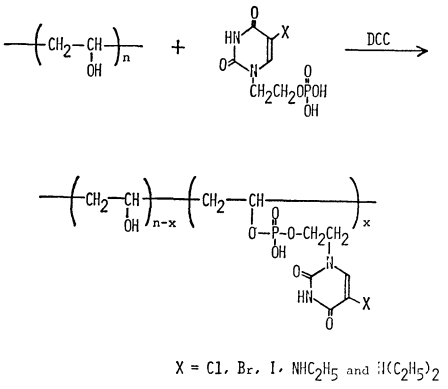
This paper deals with the syntheses of 5-substituted 1-[(2′-dihydrogenphosphato)-ethyl]-uracils and their coupling with poly(vinyl alcohol).

1-(2′-Hydroxyethyl)-uracil (I) was readily converted into 1-(2′-hydroxyethyl)-5-iodouracil (III) in good yield by refluxing with iodine in 1,4-dioxane and 0.5 M nitric acid. Reaction of compound (III) with phosphorus oxychloride in trimethyl phosphate and subsequent hydrolysis gave 1-[(2′-dihydrogenphosphato)-ethyl]-5-iodouracil (IV). Compound (IV) was also obtained by refluxing 1-[(2′-dihydrogenphosphato)ethyl]-uracil (II) with iodine in 1,4-dioxane and 0.5 M nitric acid. Bromination of (I) was carried out using the system bromine, carbon tetrachloride and 0.5 M nitric acid at room temperature. 1-[(2′-Dihydrogenphosphato)-ethyl]-5-bromouracil (V) was easily prepared by both the reactions a) of 1-(2′-hydroxyethyl)-5-bromouracil with phosphorus oxychloride and b) of the compound (II) with bromine. *N*-Ethylamino and *N,N*-diethylamino groups were introduced into uracil by the reaction of compound (V) with ethylamine and diethylamine, respectively. These reaction paths are shown in Scheme 1.



Scheme 1

As shown in Scheme 2, the condensation reaction of 5-substituted-1-[(2′-dihydrogenphosphato)-ethyl]-uracils with poly(vinyl alcohol) was carried out using *N,N′*-dicyclohexylcarbodiimide (DCC) as a dehydrating agent in refluxing dimethylformamide (DMF)-water mixture (9 : 1).



Scheme 2

The results of incorporation are given in Table 1. Nitrogen analysis showed that about half of the original hydroxy groups of poly(vinyl alcohol) was occupied by the uracil derivatives. The white powdery polymers obtained were very soluble in water and showed typical infrared absorptions at 1300—1000 cm^{-1} due to phosphoric ester. Ultraviolet absorptions were identical with those of the starting uracil derivatives.

Table 1. Condensation of 5-Substituted-1-[(2′-dihydrogenphosphato)-ethyl]-uracil (5-X-1-MOU) with PVA

X	Reaction time (hr)	Yield %	Content of 5-X-1-MOU (mol%)	UV Spectra $\lambda_{max}^{H_2O}$ (nm)
Cl	25	68	58	213, 280
Br	24	62	48	212, 282
I	20	58	42	217, 291
NHC_2H_5	25	60	52	213, 284
$N(C_2H_5)_2$	25	42	39	212, 282

Experimental

The melting points are uncorrected. The infrared (IR) spectra were obtained on a JASCO Model IR-G Spectrometer. The ultraviolet (UV) spectra were measured with a Hitachi Recording Spectrometer Model EPS-3T. Paper chromatography was carried out by the ascending technique using Toyo Roshi No 50 paper. Solvents for chromatography were a mixture of *n*-propanol, concd NH_4OH and water (6 : 2 : 2) (solvent A), and a mixture of concd NH_4OH, ethanol and water (10 : 15 : 2) (solvent B).

Poly(vinyl alcohol) used as stem polymer had a degree of polymerization of 300.

1-(2′-Hydroxyethyl)-5-iodouracil (III). Iodine 2.58 g (20 mmol) was added to a solution of 1-(2′-hydroxyethyl)-uracil (I) 1.7 g (10.9 mmol) in 0.5 M nitric acid (12 ml) and 1,4-dioxane (48 ml). The solution was heated at 100 °C for 4 hr. After the reaction mixture was allowed to cool to room temperature, the solvents were evaporated under reduced pressure. The residue was recrystallized from ethanol to give colorless prism crystals. Yield 85%; mp 197—200 °C; R_f of tlc 0.88 (solvent B); $\lambda_{max}^{H_2O}$, 217 mμ (ε=10440), 290 mμ (ε=8156); IR, 3350 (s), 1710(s), 1670(s), 1450(m), 1440(w), 1350(s), 1250(m), 1130(w), 1050(m),

3) T. Seita, K. Yamauchi, M. Kinoshita, and M. Imoto, *ibid.*, **154**, 263 (1972).

1010(s), 900(w), 600(s) cm^{-1}; Found: C, 25.54; H, 2.51; N, 9.89%. Calcd for $C_6H_7O_3N_2I$: C, 25.58; H, 2.44; N, 9.93%.

1-(2'-Hydroxyethyl)-5-bromouracil (VI). Bromine 1.2 g (15 mmol) dissolved in 8 ml of carbon tetrachloride was added to a solution of (I) 1.5 g (9.6 mmol) in a mixture of 0.5 M nitric acid (8 ml) and 1,4-dioxane (32 ml), and the solution was stirred at room temperature for 5 hr. The solvent were evaporated to dryness under reduced pressure. The residue was recrystallized from ethanol to give colorless needle crystals. Yield 87%; mp 209—211 °C; R_f of tlc 0.86 (solvent B); $\lambda_{max}^{H_2O}$, 212 mμ (ε=9251), 281 mμ (ε=8980); IR, 3350(s), 1710(s), 1670(s), 1450(m), 1340(s), 1230(w), 1050(s), 930(m), 880(w), 860(m), 750(s) cm^{-1}; Found: C, 30.51; H, 3.04; N, 11.81%. Calcd for $C_6H_7O_3N_2Br$: C, 30.64; H, 3.01; N, 11.92%.

1-[2'-Dihydrogenphosphato)-ethyl]-5-iodouracil (IV). a) 2.4 g of iodine (18.9 mmol) was added to a solution of 1.2 g of 1-[2'-dihydrogenphosphato)-ethyl]-uracil (II) (5.09 mmol) in 0.5 M nitric acid (10 ml) and 1,4-dioxane (40 ml). The procedure was similar to that for (III). Colorless powdery crystals were obtained by recrystallization from a minimal amount of absolute ethanol. Yield 69%.

b) 0.46 g of phosphorus oxychloride (3 mmol) was added at 0 °C to a suspension of 0.7 g of (III) (2.48 mmol) in trimethyl phosphate (7 ml). After stirring for 6 hr, water (3 ml) was added and stirring was continued for 1 hr. The water added was removed under reduced pressure below 50 °C, and the residue was poured into ethanol–ether mixture. The white precipitate was recrystallized from a minimal amount of absolute ethanol to give colorless powdery crystals. mp 216—217 °C; R_f of paper chromatography 0.25 (solvent A); $\lambda_{max}^{H_2O}$, 217 mμ (ε=10655), 290 mμ (ε=8196); IR, 3000(s), 1700(s), 1650(s), 1470(m), 1430(w), 1240(m), 1060(w), 1040(s), 980(s), 850(w), 660(w), 620(m) cm^{-1}; Found: C, 20.31; H, 2.21, N, 8.18%. Calcd for $C_6H_9N_2O_6PI$: C, 19.90; H, 2.23; N, 7.74%.

1-[(2'-Dihydrogenphosphato)-ethyl]-5-bromouracil (V). a) 0.5 g of (II) (2.1 mmol) was dissolved in a hot mixture of 0.5 M nitric acid (4 ml) and 1.4-dioxane (16 ml). A solution of 0.48 g of bromine (6 mmol) in carbon tetrachloride (4 ml) was added to the above solution at room temperature. After being kept at room temperature overnight, the solvents were evaporated under reduced pressure. The residue was recrystallized from ethanol–ether mixture to give colorless needle crystals. Yield 68%.

b) 0.4 g of phosphorus oxychloride (3 mmol) was added to a suspension of 0.5 g of (VI) (2.13 mmol) in trimethyl phosphate (5 ml) and treated as the synthesis of (IV)-b). Colorless needle crystals were obtained by recrystallization from ether–ethanol mixture. mp 236—237 °C; R_f of paper chromatography 0.33 (solvent A); $\lambda_{max}^{H_2O}$, 212 mμ (ε=8414), 281 mμ (ε=8090); IR, 3000(s), 1700(s), 1650(s), 1470(m), 1430(w), 1350(w), 1040(s), 980(s), 650(w), 620(m), 530(m) cm^{-1}; Found: C, 23.39; H, 2.49; N, 9.28%. Calcd for $C_6H_9N_2O_6PBr$: C, 22.88; H, 2.56; N, 8.89%.

1-[(1'-Dihydrogenphosphato)-ethyl]-5-chlorouracil (IX). 5.35 g of chlorine (7.5 mmol) dissolved in carbon tetrachloride (10 ml) was added to a solution of 1.0 g of (II) (4.2 mmol) in acetic acid (60 ml) and the solution was kept at room temperature for 1 hr. The solution was then concentrated to half the volume and refluxed for 30 min. The solvents were evaporated to dryness under reduced pressure. The residue was recrystallized from the ether–ethanol mixture to give colorless crystals. Yield 90%, mp 212—214 °C, $\lambda_{max}^{H_2O}$, 213 mμ (ε=12000), 280 mμ (ε=9800); IR, 3050(m), 1690(s), 1640(s), 1460(m), 1420(m), 1060(w), 1030(s), 980(m), 660(w), 520(m) cm^{-1}; Found: C, 26.19; H, 2.89; N, 9.92; Cl, 12.34%. Calcd for $C_6H_8N_2O_6PCl$: C, 26.64; H, 2.98; N, 10.35; Cl, 13.10%.

1-[(2'-Dihydrogenphosphato)-ethyl]-5-(N-ethylamino)-uracil (VII). A solution of 0.35 g of (V) (1.0 mmol) in 50% ethylamine (10 ml) was stirred for 24 hr at room temperature. The reaction mixture was evaporated to dryness under reduced pressure. Colorless plate crystals were obtained by recrystallization from ethanol–water mixture. Yield 58%, mp 214—216 °C, R_f of paper chromatography 0.27 (solvent A), $\lambda_{max}^{H_2O}$, 212 mμ (ε=7090), 281 mμ (ε=6986): IR 3150(s), 1710(s), 1610(m), 1440(s), 1380(w), 1340(s), 1240(m), 1010 (s), 780(m), 640(w) cm^{-1}; Found: C, 27.84; H, 5.48; N, 16.07%. Calcd for $C_8H_{14}N_3O_6P$: C, 28.25; H, 5.53; N, 16.47%.

1-[(2'-Dihydrogenphosphato)-ethyl]-5-(N,N-diethylamino)-uracil (VIII). A solution of 0.35 g of (V) (1.0 mmol) in 50% diethylamine (12 ml) was refluxed for 5 hr and then treated as in the synthesis of (VI). Yield 53%, mp 235—236 °C, R_f of paper chromatography 0.22 (solvent A), $\lambda_{max}^{H_2O}$, 212 mμ (ε=9064), 281 mμ (ε=8953); IR, 2540(s), 1680(s), 1610(m), 1500(w), 1420(m), 1340(s), 1230(s), 1150(m), 1030(s), 890(s), 640(w) cm^{-1}; Found: C, 38.51; H, 5.79; N, 13.24%. Calcd for $C_{10}H_{18}N_3O_6P$: C, 39.10; H, 5.90; N, 13.68%.

Condensation of 5-Substituted 1-[(2'-Dihydrogenphosphato)-ethyl]-uracils with Poly(vinyl alcohol). 5-Substituted 1-[(2'-dihydrogenphosphato)-ethyl]-uracil (7.7 mmol) and PVA (0.22 g) were dissolved in boiling water. To the solution was added DMF (12 ml) and the water was then evaporated to substitute the solvent for DMF. DDC (30 g) was added to the DMF solution and the mixture was refluxed for 25—25 hr. After cooling the reaction mixture was poured into a large amount of water and allowed to stand at room temperature overnight. The mixture was filtered and the filtrate was evaporated under reduced pressure to obtain a white polymer as residue. The polymer obtained was purified by column chromatography with Sephadex G-25 eluted with water.