group of pyrimidine[3] led us to attempt the synthesis of 8,2′-cyclonucleoside.

2′-*O*-Acetyl-3′-*O*-tosyl-5′-*O*-methoxycarbonyl-D-xylofuranosyl chloride[4] was condensed with 2,8-dichloroadenine chloromercury salt in refluxing xylene to give 2,8-dichloro-9-(2′-*O*-acetyl-3′-*O*-tosyl-5′-*O*-methoxycarbonyl)-β-D-xylofuranosyladenine, m.p. 160–162° (*Anal.* Calcd. for $C_{21}H_{21}O_9N_5Cl_2S$: C, 42.85; H, 3.60; N, 11.90. Found: C, 42.56; H, 3.79; N, 11.90. $\lambda_{max}^{0.1\,N\,HCl}$ 263 mμ, $\lambda_{max}^{0.1\,N\,NaOH}$ 263 mμ. Paper chromatography: R_f 0.90 (1-butanol: water = 86:14). This material was converted to its 8-mercapto derivative by the reaction with one equivalent of thiourea. Although 2-chloro-8-mercapto-9-(2′-*O*-acetyl-3′-*O*-tosyl-5′-*O*-methoxycarbonyl)-β-D-xylofuranosyladenine (I) was obtained in the form of a hard glass (yield 68.2%), evidence for its assigned structure was obtained by paper chromatography (R_f 0.90 (1-butanol: water = 86:14)) and spectrophotometric analysis ($\lambda_{max}^{0.1\,N\,HCl}$ 302, 310 mμ; λ_{max}^{EtOH} 302, 310 mμ, $\lambda_{max}^{0.1\,N\,NaOH}$ 300 mμ).

Refluxing of I in methanol containing sodium methoxide for 12 min. gave a compound having m.p. 228–229° dec. $\lambda_{max}^{0.1\,N\,HCl}$ 277 mμ, $\lambda_{max}^{0.1\,N\,NaOH}$ 277 mμ. *Anal.* Calcd. for $C_{10}H_{10}O_3N_5ClS.0.5H_2O$: C, 36.99; H, 3.38; N, 21.56. Found: C, 36.66; H, 3.82; N, 20.80. Paper chromatography [R_f 0.42 (water, pH 10.0), R_{Ad}[5]]. From spectrophotometric, chromatographic and elemental analytical data, the resulting compound was concluded to be 2-chloro-8-mercapto-8,2′-anhydro-D-xylofuranosyladenine (II) and not the presumed intermediate 2′,3′-anhydronucleoside (III). This was further supported by the fact that II has no ν_{max}^{Nujol} at 863 cm.$^{-1}$, which has been assigned to the epoxide group.[4]

The structure of II confirmed by the desulfurization of compound II with Raney nickel, which afforded 2-chloro-2′-deoxyadenosine (IV) [R_f 0.52 (water, pH 10.0), $\lambda_{max}^{0.1\,N\,HCl}$ 265, $\lambda_{max}^{0.1\,N\,NaOH}$ 265 mμ]. Hydrogenation of compound IV over palladized charcoal as a catalyst gave 2′-deoxyadenosine [R_f 0.35 (1-butanol–water = 86:14), 0.50 (water, pH 10.0); $\lambda_{max}^{0.1\,N\,HCl}$ 258, $\lambda_{max}^{0.1\,N\,NaOH}$ 260 mμ]. This material was compared directly with the authentic 2′-deoxyadenosine[6] and 3′-deoxyadenosine[7]

Sample	Hydrolysis product	R_f[a]	Spray I[b]	Spray II[c]
Natural 2′-deoxy-adenosine	2-Deoxyribose	0.21	Pink ++	Pink +
Synthetic 2′-deoxy-adenosine	2-Deoxyribose	.21	++	+
3′-deoxyadenosine	3-Deoxyribose	.41	+	+++

[a] Paper chromatography, 1-butanol–acetic acid–water = 4:1:5, after equilibration overnight, upper layer was used. [b] Cysteine–sulfuric acid reagent: J. G. Buchanan, *Nature*, **168**, 1091 (1951). [c] Aniline hydrogen phthalate reagent: S. M. Partridge, *Nature*, **164**, 443(1949).

These results clearly show that the resulting deoxynucleoside is 2′-deoxyadenosine and not the 3′-isomer. Thus it must be concluded that the 8,2′-cyclic bond was formed by the nucleophilic attack of the 8-mercapto group on the 2′-carbon atom of the epoxide linkage in III.

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(5) R_{ad} stands for a value obtained from R_f divided by R_f of adenosine.

(6) Purchased from Sigma Chemical Co. St. Louis, Mo.

(7) Unpublished experiments by A. Yamazaki and M. Ikehara.

It is hoped that 8,2′-cyclonucleoside will be useful in transforming the sugar moiety of purine nucleosides in a manner similar to that which has been used so successfully in the pyrimidine series.[8] Experiments along this line are now in progress in our Laboratory.

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Evidence for Chain Transfer in the Autoxidation of Hydrocarbons Retarded by Phenol

Sir:

Peroxy radical–antioxidant complexes have been postulated by Hammond, *et al.*,[1] to be intermediates in the inhibition of the autoxidation of hydrocarbons. Despite numerous attempts to establish the existence of such complexes,[2–4] no direct evidence has yet been obtained for their existence and, at the same time, no plausible alternative mechanism has yet been proposed.

The postulation of these complexes was based on the kinetic evidence that the rate of oxygen absorption during the oxidation of tetralin in the presence of phenol or N-methyl aniline followed the kinetic relation

$$-\frac{dO_2}{dt} \propto \frac{[Ri]^{1/2}}{[\text{inhibitor}]^{1/2}}$$

where R_i is the rate of radical production from azobis-(2-methyl propionitrile) (AIBN). This postulation, however, assumed that the rate was first order with respect to hydrocarbon concentration.

In the present work it has now been demonstrated that the dependence of the rate of oxygen absorption with respect to the concentration of the hydrocarbon is not first order. Data are presented in Fig. 1 for the initial rate of oxygen absorption by chlorobenzene solutions of phenol, AIBN, and tetralin. When phenol was absent, the rate was first order with respect to hydrocarbon concentration, demonstrating that the rate of formation of radicals from AIBN did not vary with hydrocarbon concentration. However, when phenol was added, the rate became $^3/_2$ order with respect to the hydrocarbon. Similar results were obtained using 9,10-dihydroanthracene as the substrate. Thus

$$-\frac{dO_2}{dt} \propto \frac{[R_i]^{1/2}\,[RH]^{3/2}}{[\text{phenol}]^{1/2}}$$

The kinetic chain lengths in these experiments ranged from 4 to 30.

A mechanism which can account for the observed kinetics follows

$$R' - N = N - R' \xrightarrow{k_1} 2aR'\cdot + N_2 \quad R_i = 2ak_1[AIBN] \quad (1)$$

$$R'\cdot + O_2 \xrightarrow{k_2} R'O_2\cdot \quad (2)$$

$$R'O_2\cdot \text{ (or } RO_2\cdot) + RH \xrightarrow{k_3} R'O_2H + R\cdot \quad (3)$$

$$R\cdot + O_2 \xrightarrow{k_4} RO_2\cdot \quad (4)$$

$$RO_2\cdot + AOH \xrightarrow{k_5} RO_2H + AO\cdot \quad (5)$$

$$AO\cdot + RH \xrightarrow{k_6} AOH + R\cdot \quad (6)$$

$$RO_2\cdot + AO\cdot \xrightarrow{k_7} \text{inert products} \quad (7)$$

where AOH is phenol, AO· is the phenoxy radical and RH is tetralin. If one makes the usual steady state

(1) G. S. Hammond, C. S. Boozer, C. E. Hamilton, and J. N. Sen, *J. Am. Chem. Soc.*, **77**, 3238 (1955).

(2) K. U. Ingold, *Chem. Rev.*, **61**, 563 (1961).

(3) J. R. Thomas and C. A. Tolman, *J. Am. Chem. Soc.*, **84**, 2930 (1962).

(4) J. R. Thomas, *ibid.*, **85**, 591, 593 (1963).

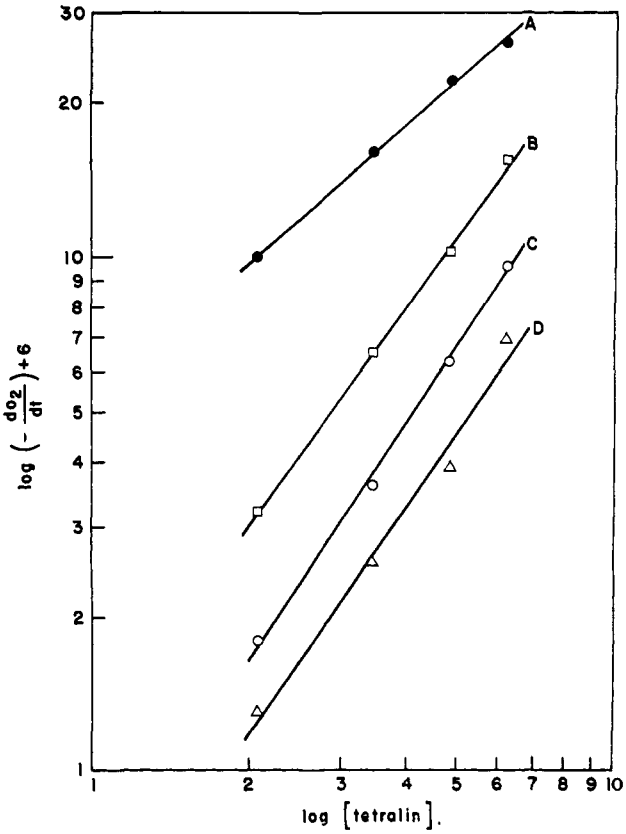

Fig. 1.---Dependence of rate of oxygen absorption on tetralin concentration at 60° and 37 × 10^{-3} *M* AIBN: A, no phenol, slope = 0.90; B, 7.6 × 10^{-4} *M* phenol, slope = 1.42; C, 23 × 10^{-4} *M* phenol, slope = 1.53; D, 38 × 10^{-4} *M* phenol, slope = 1.48.

assumptions and the further approximations

$$k_3[\mathrm{RH}] \gg k_5[\mathrm{AOH}]$$

and

$$k_6[\mathrm{RH}] \gg k_7[\mathrm{RO_2}\cdot],$$

then

$$-\frac{d\mathrm{O_2}}{dt} = k_3\left(\frac{k_6R_i}{2k_5k_7}\right)^{1/2}\frac{[\mathrm{RH}]^{3/2}}{[\mathrm{phenol}]^{1/2}}$$

Thus, the kinetic expression obtained for the phenol–tetralin system is consistent with a mechanism in which chain transfer,[5,6] reaction 6, is of considerable importance, with no necessity of invoking a peroxy radical–inhibitor complex. Other inhibitors whose kinetic behavior has been interpreted in terms of "complex formation" (*viz.*, N-methylaniline,[1] diphenylamine,[3] trialkylamines[4]) are substances which, like phenol, would give relatively unreactive monofunctional radicals and which therefore could restart chains by a similar chain transfer mechanism.

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The Selective Acetylation of Terminal Hydroxyl Groups in Deoxyribo-oligonucleotides[1]

Sir:

As a part of work on the sequential analysis of nucleic acids, recently, methods for the labeling of terminal phosphomonoester groups in polynucleotides have been reported from this Laboratory.[2,3] The present communication describes a method for the selective acetylation of the free hydroxyl groups, on the terminal nucleosides in deoxyribopolynucleotides. The approach thus promises to be complementary to those developed previously[2,3] for the labeling of end groups in nucleic acids.

TABLE I

RESULTS OF ACETYLATION OF 3′-HYDROXYL GROUPS IN MONO- AND OLIGONUCLEOTIDES

Compound[4]	Concentration (μmole/ml.)	Ac_2O (mmoles)	Yield	Assay
pT	0.72	1	63	1
d-pA	.70	1	85	1
d-pG	.70	1	88	1
d-pC	.70	1	71	1
		2	81	1
		3	96	1
pTpTpTpT	.18	1	55	1, 2
		3	70	1, 2
d-pApApApA	.18	1	75	1, 2
		3	80	1
d-pCpCpCpC	.18	1	73	2
		3	87	2
d-pApApApApApApA	.035	1	62	2

A solution of the mononucleotide or oligonucleotide (Table I) (0.07–1.4 μmoles of the sodium or ammonium salt) in water (2.0 ml.) was treated with acetic anhydride (0.1–0.3 ml.) at room temperature, the mixture being stirred by a magnetic stirrer. Acetic anhydride was added in portions of 0.01 ml., the pH of 7 maintained by the continuous addition of 4 *N* sodium hydroxide from a microsyringe. After the completion of the addition (about 15 min.), the nucleotidic material was freed from sodium acetate either by extensive dialysis against water (for oligonucleotides) or by passage of the total solution through a column of pyridinium Dowex-50 ion-exchange resin and lyophilization of the resulting solution. Two types of assays were used for following the extent of the acetylation reaction. In assay 1, the mixture was chromatographed in the solvent *n*-butyl alcohol–acetic acid–water (5–2–3) or ethyl alcohol–0.5 *M* ammonium acetate (pH 3.8) (7–3, v./v.). In the case of mononucleotides and the tetranucleotides pTpTpTpT[4] and *d*-pApApApA,[4] the 3′-*O*-acetyl derivatives were clearly resolved from the starting materials. The assay 2 involved incubation of the mixture with the *Escherichia coli* phosphodiesterase[5] in order to degrade the unchanged oligonucleotide to mono- and dinucleotides and to determine the acetylated product by paper chromatography. It has been demonstrated recently that acetylation of the 3′-hydroxyl group confers complete resistance on the oligonucleotide towards this enzyme.[6] The results are given in Table I. In general, the yields were high and probably can be increased further. It is worth noting that the yields in the case of the tetranucleotides and the one heptanucleotide studied showed no decrease.

Previous experience[7] of acylation of nucleotides in

(1) This work has been supported by grants from the National Cancer Institute of the National Institutes of Health, The National Science Foundation, Washington and The Life Insurance Medical Research Fund, New York, N. Y.

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