There was isolated from another condensation reaction a small amount of material which crystallized from ethyl acetate as tiny colorless needles, m.p. 288.5–289°. Satisfactory purification was not possible because of the small quantity obtained, but the material appeared to be mainly **1,14-dihydroxytricyclo[13.11.1.1^{2,4}]octacosane-27,28-dione.**

Anal. Calcd. for $C_{28}H_{48}O_4$: C, 74.95; H, 10.78. Found: C, 74.54, 74.22; H, 10.90, 10.54.

The infrared absorption spectrum (1% in chloroform) showed maxima at 3456 (OH), 1706 (C=O) and a weak band at 1605 cm.$^{-1}$ (C=C?).

Preparation of 3-Benzyl-1,2-cyclotridecanedione.—Diethyl α-benzylbrassylate was prepared *via* a malonic ester synthesis, using ethyl 11-bromohendecanoate and diethyl benzylmalonate, in 51% yield, b.p. 176–178° (0.2 mm.), n^{19}D 1.4802, infrared C=O maximum at 1737 cm.$^{-1}$.

Anal. Calcd. for $C_{24}H_{38}O_4$: C, 73.80; H, 9.81. Found: C, 73.77; H, 9.84.

The dianilide of α-benzylbrassylic acid formed colorless crystals from methanol, m.p. 145–146°.

Anal. Calcd. for $C_{32}H_{40}N_2O_2$: C, 79.30; H, 8.32; N, 5.78. Found: C, 79.26; H, 8.22; N, 5.91.

The acyloin condensation of diethyl α-benzylbrassylate was carried out in the usual manner and the crude product was oxidized directly with cupric acetate in acetic acid[29] to **3-benzyl-1,2-cyclotridecanedione,** b.p. 139–140°, n^{20}D 1.5244; infrared C=O maximum (5% chloroform) at 1708 cm.$^{-1}$; ultraviolet maximum (cyclohexane) at 448 (43) mμ.

Anal. Calcd. for $C_{20}H_{28}O_2$: C, 79.95; H, 9.39. Found: C, 79.70; H, 9.44.

The quinoxaline of 3-benzyl-1,2-cyclotridecanedione was obtained by heating the diketone with 10% molar excess of *o*-phenylenediamine in absolute ethanol under reflux for 2 hours and was purified by recrystallization from 95% ethanol, light yellow crystals, m.p. 80.5–82°.

Anal. Calcd. for $C_{26}H_{32}N_2$: C, 83.82; H, 8.66; N, 7.52. Found: C, 83.71; H, 8.68; N, 7.61.

Preparation of 4,4-Dimethyl-2,3-pentanedione. 4-Isonitroso-2,2-dimethyl-3-pentanone.—An ether solution (100 g.) containing 0.55 mole of hydrogen chloride was added to a solution of 57 g. (0.5 mole) of 2,2-dimethyl-3-pentanone[38] in 570 g. of anhydrous benzene. The resulting solution was stirred and cooled externally. When the internal temperature was −8°, a solution of 57 g. (0.55 mole) of *n*-butyl nitrite[39] in 280 g. of absolute ether was added during 1.5 hours while stirring and maintaining at −8 to −10°. The deep red solution was kept in a refrigerator for 2.5 days, after which the color was light orange. The reaction mixture was washed with three 500-ml. portions of ice-water. The organic layer was extracted with two 500-ml. portions of cold 1 *N* sodium hydroxide. The combined cold alkaline extracts were carefully neutralized to *p*H 7 with cold dilute sulfuric acid. The precipitated product was collected, dried and recrystallized from hexane as colorless needles, m.p. 110.5–111.5°, yield 59 g. (82%); infrared maxima at 3370 (OH), 1672 (conj. C=O) and 1632 cm.$^{-1}$ (C=N).

Anal. Calcd. for $C_7H_{13}NO_2$: C, 58.72; H, 9.15; N, 9.78. Found: C, 58.88; H, 8.97; N, 9.62.

4,4-Dimethyl-2,3-pentanedione dioxime was prepared from the isonitrosoketone and hydroxylamine hydrochloride in ethanol–pyridine and recrystallized from aqueous ethanol as colorless needles, m.p. 183.5–184° (reported[40] 182.5°).

Anal. Calcd. for $C_7H_{14}N_2O_2$: C, 53.14; H, 8.92; N, 17.71. Found: C, 53.44; H, 9.22; N, 17.58.

4,4-Dimethyl-2,3-pentanedione was obtained from 4-isonitroso-2,2-dimethyl-3-pentanone by the method of Fréon and Ser,[41] using concentrated hydrochloric acid, with formalin as the hydroxylamine acceptor; yield 26%, b.p. 125–126°, n^{20}D 1.4047; infrared C=O band centered at 1710 cm.$^{-1}$.

Anal. Calcd. for $C_7H_{12}O_2$: C, 65.59; H, 9.44. Found: C, 65.97; H, 9.54.

The dioxime was identical with the hydroxylamine condensation product from 4-isonitroso-2,2-dimethyl-3-pentanone. The **bis-2,4-dinitrophenylhydrazone** was obtained as yellow-orange crystals from aqueous dimethylformamide, m.p. 220.5–221.5° (reported[40] 214–215°).

Condensation of Benzaldehyde with 4,4-Dimethyl-2,3-pentanedione.—The condensation of 4,4-dimethyl-2,3-pentanedione and benzaldehyde was carried out in the presence of a trace of piperidine in refluxing ethanol solution to give **1-phenyl-5,5-dimethyl-1-hexene-3,4-dione** in 34% yield, b.p. 66° (0.03 mm.), n^{19}D 1.5641.

Anal. Calcd. for $C_{14}H_{16}O_2$: C, 77.75; H, 7.46. Found: C, 78.01; H, 7.36.

(38) F. C. Whitmore, C. I. Noll and V. C. Meunier, THIS JOURNAL, **61**, 683 (1939).

(39) E. Karg, *Arch. Pharm.*, **282**, 49 (1944).

(40) E. B. Reid and R. B. Fortenbaugh, *J. Org. Chem.*, **16**, 33 (1951).

(41) P. Fréon and S. Ser, *Compt. rend.*, **222**, 447 (1946).

URBANA, ILLINOIS

[CONTRIBUTION FROM THE GEORGE HERBERT JONES LABORATORY, UNIVERSITY OF CHICAGO]

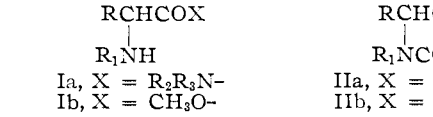
Some Reactions of Amino Acid N-Carboxy Anhydrides

BY KENNETH D. KOPPLE

RECEIVED JUNE 1, 1957

Primary amino acid N-carboxy anhydrides (NCA's) react with amines and sodium methoxide to yield carbamic acid derivatives. It is suggested that these derivatives arise *via* intermediate α-isocyanato acids. Evidence is adduced to demonstrate that under basic conditions NCA's can be converted to such intermediates and that isocyanato acids are likely intermediates in certain NCA polymerization reactions. An approximate equilibrium constant for base-catalyzed dissociation in dioxane of *p*-nitrophenyl N-phenylcarbamate into the corresponding phenol and phenyl isocyanate has been determined.

In a recent paper, it was reported that amino acid N-carboxy anhydrides (NCA's) react with excess of primary and secondary amines, yielding



Ia, X = R_2R_3N-
Ib, X = CH_3O-

IIa, X = R_2R_3N-
IIb, X = CH_3O-

both α-amino amides (Ia) and α-ureido acids (IIa). With a given anhydride, the ratio of the two products is dependent on the amine.[1] Extension of this study to include reaction with sodium methoxide has led to the results which are cited in Table I.

It was suggested previously that Ia and IIa are products of nucleophilic addition to the 5- and 2-carbonyl groups, respectively, of the anhydride.[1]

(1) K. D. Kopple, THIS JOURNAL, **79**, 662 (1957).

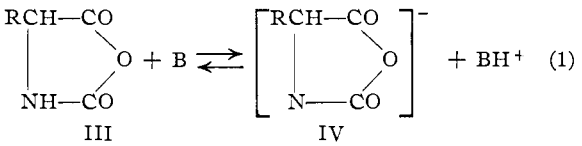
TABLE I

PERCENTAGE YIELD OF II IN REACTION OF NCA'S WITH *t*-BUTYLAMINE AND METHOXIDE ION

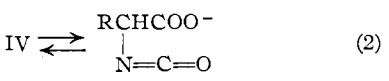
NCA	t-$C_4H_9NH_2$[a]	CH_3O^-[b]
Glycine	90[c]	44[d]
Sarcosine	0[c]	0
α-Aminoisobutyric	80	42

[a] Large excess of amine. [b] One equivalent of sodium methoxide plus excess methanol. [c] Reference 1. [d] Similar results have been reported for γ-benzyl glutamate NCA by Blout and Karlson, reference 7a.

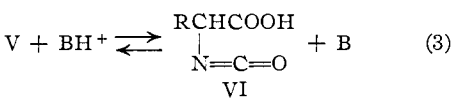
Nucleophilic attack at the 5-carbonyl is an acceptable reaction path[2]; but the absence of IIb (R_1 = CH_3) from among the reaction products of sarcosine NCA with the powerfully nucleophilic methoxide ion makes it appear unlikely that similar reaction occurs at the 2-carbonyl. Since, as Table I indicates, urethans or ureas are not produced at all from sarcosine NCA, their formation presumably requires replaceable hydrogen at the anhydride nitrogen atom. A reaction scheme which embodies this requirement involves the reactions shown in equations 1–3, followed by reaction of V or VI with



III IV (1)



V (2)



VI (3)

alcohol (alkoxide) or amine in the usual manner of isocyanates. Formation of carbamic acid derivatives thus would depend on successful competition of these reactions with direct reaction of amine or alkoxide at the 5-carbonyl of III. Factors favoring the formation of isocyanate, and ultimately of carbamic acid derivatives, would include increased basicity of the reaction medium and decreased ability of the nucleophilic reagent to add to the 5-carbonyl group (*e.g.*, for steric reasons). The data of Table I and of the previous report[1] fit this scheme.[3]

It is difficult to show the presence of V or VI in solutions of N-carboxy anhydrides, since conditions appropriate to such demonstration lead to rapid polymerization. In model systems, however, a similar reaction readily is observed. The infrared spectra, in dioxane solution, of *p*-nitrophenyl N-phenylcarbamate and of 2,4-dinitrophenyl N-phenylcarbamate are much as expected, although they exhibit high C=O stretching frequencies (1750 and 1760 cm.$^{-1}$, respectively). On addition of a tertiary amine to the dioxane solution it is possible to observe the equilibrium of equation 4, as shown by a decrease in the intensity

$$\mathrm{ArOCONHC_6H_5} \underset{\text{base}}{\overset{K}{\rightleftharpoons}} \mathrm{ArOH} + \mathrm{C_6H_5NCO} \qquad (4)$$

of absorption due to urethan carbonyl and the appearance of phenyl isocyanate absorption at 2230 cm.$^{-1}$. For the *p*-nitrophenyl compound, K at room temperature is about 2×10^{-3}. A dioxane solution of the dinitrophenyl compound, on addition of tertiary amine, precipitates the dinitrophenolate salt. Mukaiyama and co-workers[4] have already suggested this dissociation of urethans on the basis of kinetic studies of the reaction

$$\mathrm{ROCONHR' + R''NH_2(solvent)} \longrightarrow \mathrm{R''NHCONHR' + ROH}$$

at elevated temperatures.

Infrared spectra of solutions of the two urethans in dimethylformamide 1.0 M in lithium chloride also exhibit moderate isocyanate absorption, while solutions in pure dimethylformamide are only weakly absorbing.

Amino acid N-carboxy anhydrides may be viewed as cyclic carbamates in which the alkoxy moiety has been replaced by a carboxyl group. Since the carboxyl group is of comparable acidity to 2,4-dinitrophenol, base-catalyzed ring opening of NCA's is also to be expected, although to a lesser extent than dissociation of the acyclic urethans. That N-carboxy anhydrides (III) are the stable form of α-isocyanato acids (VI) is indicated by the Curtius synthesis of NCA's from the half azides of substituted malonic acids.[5] In this, as in other acyl azide decompositions, an isocyanate is probably an intermediate.

It has proved possible to demonstrate reproducibly the presence of an isocyanate-containing species derived from an NCA, although under non-equilibrium conditions. If a solution of the N-carboxy anhydride of α-aminoisobutyric acid in dioxane be shaken briefly with excess sodium hydride, hydrogen is evolved. The infrared spectrum of the supernatant liquid then exhibits isocyanate absorption which decreases with time. (The choice of anhydride for this experiment was suggested by the observation that α-aminoisobutyric acid NCA polymerizes much more slowly under these and other strongly basic conditions than do the more common amino acid NCA's, and by the report that α-isocyanato esters react relatively slowly with acids.[10])

The synthesis of poly-α-amino acids from NCA's in a reaction catalyzed by salts of weak acids,[6a] by tertiary amines[6b] or by solutions of inorganic salts in dimethylformamide[6c] has been reported. Ballard, Bamford and Weymouth have examined both the rates and products of these polymeriza-

(2) C. H. Bamford, A. Elliott and W. E. Hanby, "Synthetic Polypeptides," Academic Press, Inc., New York, N. Y., 1956, pp. 62–78.

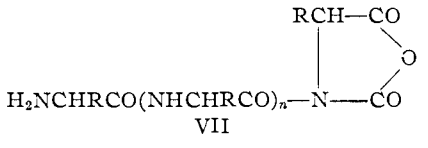
(3) P. D. Bartlett and R. H. Jones, THIS JOURNAL, **79**, 2153 (1957), report general base catalysis of the hydrolysis of NCA's in aqueous solution. This observation is consistent with the scheme above but implies that proton removal from the NCA is the rate-determining step of the hydrolytic sequence. It is not obvious, however, that proton removal is rate determining in the reactions discussed in the present report.

(4) T. Mukaiyama and M. Iwanami, *ibid.*, **79**, 73 (1957), and earlier papers.

(5) T. Curtius and W. Sieber, *Ber.*, **54**, 1430 (1921); **55**, 1543 (1922).

(6) (a) D. G. H. Ballard and C. H. Bamford, Symposium on Peptide Chemistry, Special Publication No. 2, The Chemical Society, London, 1955, p. 25; (b) D. G. H. Ballard and C. H. Bamford, *J. Chem. Soc.*, 381 (1956); (c) D. G. H. Ballard, C. H. Bamford and F. J. Weymouth, *Proc. Roy. Soc. (London)*. **A227**, 155 (1955); (d) ref. 2, p. 90.

tions and have shown that, in contrast to the polymerization initiated by primary amines, the reactions mentioned above do not proceed with sarcosine NCA. Further, the products of such tertiary base or salt-catalyzed reactions are chiefly cyclic peptides (with the exclusion of cyclic dipeptides) and hydantoin-3-acetic acids. On the basis of the observed products together with an analysis of rate data, these authors suggest the equilibrium of equation 1 as a first step. Polymerization, in this view, is then thought to occur *via* a series of intermediates VII, which grow by addition of IV to the anhydride terminus or by "normal"[2] propagation

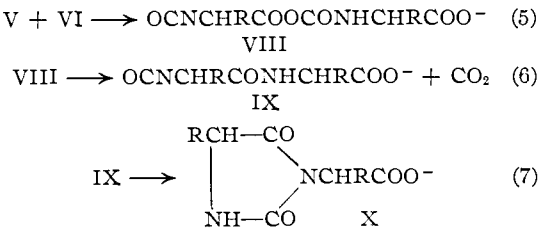


VII

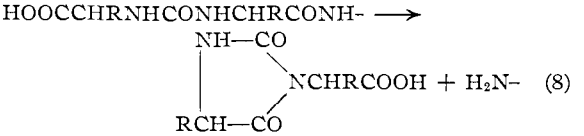
at the amino end. Cyclic peptides are logical products where $n = 0$; hydantoin-3-acetic acids may arise from the two-residue intermediate ($n = 0$). The absence of diketopiperazines is explained on steric grounds.

Alkoxide-initiated polymerization of N-carboxy anhydrides[7a,b] has been considered to proceed in a fashion similar to the above.[6d]

The high acidity required of NCA's for the scheme just described[6a,b] seems improbable to this author. An alternative reaction sequence involving isocyanate intermediates is suggested by the observations reported in this paper.[8] Equations 5–7 indicate a more likely path by which polymerization may occur.



Intermediate IX is capable of growing by reaction of carboxyl with isocyanate, combining thus with V, VI or with larger chain fragments similar to itself. It can also be expected to form a hydantoin-3-acetic acid, but formation of diketopiperazine would require an improbable eight-membered ring intermediate. Higher homologs of IX can be expected to yield cyclic peptides plus peptides with a hydantoin residue at what would have been the amino terminus. The identification of hydantoin residues at the end of polymer chains would therefore provide a means of distinguishing this scheme from that of Ballard, Bamford and Weymouth. Sela and Berger[9] have reported the isolation of hydantoin-3-acetic acids on partial hydrolysis of polymers from (non-tertiary) amine-initiated reactions. They consider these to arise, upon acid hydrolysis, from urea derivatives formed by "wrong way" addition of the growing chain to an NCA molecule (equation 8).



It is probable, however, that some hydantoin groups were hydantoin end groups in the polymer.

The addition of carboxylic acids to isocyanates to form amides and carbon dioxide, *via* unstable mixed carbamic–carboxylic anhydrides, is an established reaction which has been shown to be a practical one for the synthesis of simple peptides.[10] Good yields of protected dipeptides can be obtained by heating, in an inert solvent, an α-isocyanato ester with an acylamino acid. If pyridine is used as the solvent, the reaction is facilitated. Of those isocyanato esters studied, that derived from acetic acid reacted most rapidly, and that from isobutyric acid least.[10] This qualitative observation is in accord with the slow salt- and tertiary base-catalyzed polymerization of α-aminoisobutyric NCA. (In two hours at room temperature, no spectroscopically detectable peptide was formed in solutions of α-aminoisobutyric NCA in 0.6 *M* lithium chloride in dimethylformamide or in 0.3 *M* trimethylamine in dioxane.)

Polymerization of the type represented in equations 5 and 6 would be favored by the same factors which facilitate formation of carbamic acid derivatives from NCA's. Therefore, the reagents which, when used in excess, afford high yields of II, can be expected, in lower concentrations, to initiate such polymerization. However, these reagents are able to react with isocyanate groups to form ureas, and to react with NCA's to form aminoacyl derivatives which can react with isocyanates to form ureas. In addition, they can initiate "normal" polymerization. These processes, and others of similar nature, will act as chain-breaking reactions. In this connection, the results of Blout and Karlson are illustrative.[7a] These workers initiated the polymerization of γ-benzylglutamic ester NCA with 0.2 *M* equivalent of various bases in dioxane solution. The molecular weight of the product polymer (determined by viscosity measurements) was observed to decrease in the order triethylamine > diethylamine > sodium methoxide (in methanol) > hexylamine. This sequence parallels the decreasing tendency of the initiators to form carbamate derivatives when in high concentration relative to the NCA, as reported in this paper and reference 1.

Polymerization of N-carboxy anhydrides, when initiated by anything but an unhindered amine of only moderate basicity (*i.e.*, an amino acid amide), thus probably combines the "normal" reaction of NCA's at the amino end of a growing peptide chain with the reactions of NCA's in the isocyanato acid form. It seems reasonable to assume that thermal polymerization of these monomers also proceeds *via* rearrangement to the isocyanate derivative.

Other amino acid derivatives are known to yield polypeptides on heating alone, in dioxane or in

(7) (a) E. R. Blout and R. H. Karlson, THIS JOURNAL, **78**, 941 (1956); (b) E. R. Blout and M. Idelson, *ibid.*, **78**, 3857 (1956).

(8) Formation of polypeptides from isocyanato acids has been discussed by C. D. Hurd, C. M. Buess and L. Bauer, *J. Org. Chem.*, **17**, 865 (1952).

(9) M. Sela and A. Berger, THIS JOURNAL, **77**, 1893 (1955).

(10) S. Goldschmidt and M. Wick, *Ann.*, **575**, 217 (1952).

inert solvents containing traces of pyridine. Among these are the N-phenylthiocarbonylamino acids[11] and various substituted N-phenoxycarbonylamino acids.[12] In view of the ready dissociation of similar urethans to isocyanates it would seem that these monomers also function as sources of isocyanato acids. It is of interest to note that *o*-nitrophenoxycarbonyl-DL-alanine polymerizes more readily than the *p*-methylphenoxy analog[12]; this relationship would be expected if dissociation to isocyanate and phenol is a phase of the over-all reaction.

Acknowledgment.—This work was supported in part by the National Science Foundation, Grant No. G 2640. The author is indebted to Mrs. Danute Nitecki for her assistance in some of the experimental work, and to Dr. R. A. Clement for helpful discussions.

Experimental[13]

N-(N'-*t*-Butylcarbamyl)-α-aminoisobutyric Acid.—This substance was obtained in 77% yield by reaction of α-aminoisobutyric acid NCA with excess *t*-butylamine at 0° in the manner described previously for DL-phenylalanine NCA.[1] Recrystallization from ethyl acetate–petroleum ether afforded an analytical sample, m.p. 146–147° dec.

Anal. Calcd. for $C_9H_{18}N_2O_3$: C, 53.44; H, 8.97; N, 13.85. Found: C, 53.65; H, 9.06; N, 13.71.

Reaction of NCA's with Sodium Methoxide.—To the anhydride (1.0 g.), suspended or dissolved in 10 ml. of dioxane, was added one equivalent of 5.0 *M* sodium methoxide in methanol. Slight evolution of heat ensued. The reaction mixture was allowed to remain at room temperature for 18 hr. In the case of glycine and sarcosine NCA's a gel was formed rapidly, but with α-aminoisobutyric acid NCA as reactant, the solution remained clear. Each of the three reaction mixtures was worked up to isolate the corresponding N-carbomethoxyamino acid by the use of a cation exchanger.[1]

From glycine NCA there was obtained 580 mg. (44%) of carbomethoxyglycine, m.p. 95–96°.[14] There was also formed a considerable but undetermined amount of insoluble polyglycine.

From sarcosine NCA was obtained only 225 mg. (36%) of sarcosine anhydride, m.p. 145–146°.[15] No carbomethoxysarcosine was found, and no attempt was made to isolate sarcosine methyl ester.

From α-aminoisobutyric acid NCA was isolated 520 mg. (42%) of the corresponding N-carbomethoxy derivative. Recrystallization from ethyl acetate–petroleum ether led to an analytical sample, m.p. 158–159°.

Anal. Calcd. for $C_6H_{11}NO_4$: C, 44.71; H, 6.88; N, 8.69. Found: C, 44.89; H, 6.88; N, 8.49.

2,4-Dinitrophenyl-N-Phenylcarbamate.—A standard procedure[16] was used for the reaction of dinitrophenol and phenyl isocyanate. Careful exclusion of moisture was necessary both in carrying out the addition reaction and in recrystallization of the product. Two crystallizations from ethyl acetate–petroleum ether yielded a product of m.p. 106–107°.

Anal. Calcd. for $C_{13}H_9N_3O_6$: C, 51.49; H, 2.99. Found: C, 51.63; H, 3.25.

On contact with water, the pale yellow crystalline urethan readily decomposed with regeneration of 2,4-dinitrophenol.

Dissociation of Carbamates.—2,4-Dinitrophenyl-N-phenylcarbamate prepared as above and *p*-nitrophenyl-N-phenylcarbamate, m.p. 149–150°,[17] were used in these experiments.

Standard solutions in pure, dry dioxane of phenyl isocyanate and of *p*-nitrophenol plus 0.05 equivalent of trimethylamine (or pyridine) were mixed, diluted as desired with dioxane and allowed to remain at room temperature (20°) for 3 days. Similarly, standard solutions of *p*-nitrophenyl-N-phenylcarbamate, also containing 0.05 equivalent of base, were diluted and stored 3 days at room temperature. All solutions were made up and diluted in carefully dried serum bottles or 100-ml. volumetric flasks which were capped with rubber disks tightly held to the mouth of the vessel by crimped aluminum seals.[18] Transfer and dilution of solutions was accomplished by use of calibrated hypodermic syringes.

Infrared spectra of the solutions were measured, after the equilibration period, in 0.104-mm. sodium chloride cells on a Perkin–Elmer model 21 infrared spectrophotometer. Attention was focused on absorption due to phenyl isocyanate (2250 cm.$^{-1}$, ϵ 1020 at concentrations below 0.06 *M*) and the carbamate (1750 cm.$^{-1}$, ϵ 740 at concentrations below 0.06 *M*). Concentrations of these species were determined by use of plots of absorbance *vs.* concentration. Experiments using pyridine or trimethylamine as catalyst and initial reactant concentrations of 0.1 *M* to 0.02 *M* all yielded values of the dissociation constant between 1×10^{-3} and 3×10^{-3}. No correction was made for the interaction of phenol and the amine catalyst.

Although a similar series of experiments was not carried out using 2,4-dinitrophenyl N-phenylcarbamate, rough measurements indicate a dissociation constant about 100 times greater than for the *p*-nitro analog. An immediate yellow precipitate formed on addition of excess trimethylamine in dioxane to a solution of the dinitro derivative in the same solvent. No dissociation was observed with ethyl-N-phenylcarbamate.

Since dimethylformamide absorbs strongly in the 1700 cm.$^{-1}$ region of the infrared, dissociation constants for the carbamates in it were not obtained; 10% (w./v.) solutions of the nitrophenylcarbamates in dimethylformamide exhibited isocyanate absorption indicating 5–10% dissociation. The absorbance of this band was 3–5 times greater when the dimethylformamide solutions contained 1.0 *M* lithium chloride.

Dissociation of an N-Carboxy Anhydride.—Sodium hydride (Metal Hydrides, Inc., Beverly, Mass.) (30 mg.) was sealed in a serum bottle by means of the rubber caps and aluminum seals already mentioned. To this bottle was added, using a syringe, 1 ml. of pure, dry dioxane containing 100 mg. of α-aminoisobutyric acid NCA[19] (approximately 0.75 *M*). Hydrogen evolution began at once; a portion of the supernatant liquid was withdrawn after one minute and its infrared spectrum recorded as a function of time. No detectable peptide or carbon dioxide absorption was noted. At 2230 cm.$^{-1}$ there appeared absorption corresponding to 0.014 *M* isocyanate (if ϵ be taken as 1000). After 5 minutes at room temperature, this absorption corresponded to 0.011 *M*, and after 20 minutes to about 0.003 *M*. If the supernatant was allowed to remain in contact with the solid hydride, isocyanate absorption decreased more slowly, corresponding at 25 minutes to a concentration of about 0.007 *M*. In this last mentioned spectrum the NCA carbonyl absorption had decreased markedly and peptide bands at 1620 and 1500 cm.$^{-1}$ were evident. After 90 minutes, the reaction mixture set to a gel.

When L-leucine NCA was used in similar experiments no isocyanate absorption appeared, although formation of peptide did occur.

Infrared Examination of Polymerizing NCA's.—Infrared examinations of several polymerizing systems at temperatures from −78° [20] to 25° were carried out. The NCA's studied included α-aminoisobutyric acid, DL-alanine and L-leucine NCA's. Polymerization was initiated by sodium methoxide or trimethylamine in tetrahydrofuran–dioxane or by lith-

(11) J. Noguchi and T. Hayakawa, THIS JOURNAL, **76**, 2846 (1954); J. Noguchi, *et al.*, *J. Chem. Soc., Japan*, **75**, 639 (1954).

(12) Y. Iwakura, Y. Ishizuka and T. Saito, *ibid.*, **76**, 1108 (1955).

(13) Thanks are due Mr. William Saschek for the microanalyses reported in this paper.

(14) H. Leuchs, *Ber.*, **39**, 859 (1906).

(15) F. Mylius, *ibid.*, **17**, 287 (1884).

(16) R. L. Shriner, R. C. Fuson and D. Y. Curtin, "The Systematic Identification of Organic Compounds," John Wiley and Sons, Inc., New York, N. Y., 1956, p. 265.

(17) M. Busch, G. Blume and E. Pungs, *J. prakt. Chem.*, [2] **79**, 533 (1909).

(18) The rubber stoppers and aluminum seals, along with the serum bottles, were obtained from the T. C. Wheaton Co., Millville, N. J.

(19) A. C. Farthing, *J. Chem. Soc.*, 3216 (1950).

(20) An infrared cell, with silver chloride windows, capable of being filled with solution at Dry Ice temperature and maintained there, was constructed in this Laboratory by Mr. Hans Riehm and Mr. C. C. van Hespen.

ium chloride in dimethylformamide. In the region examined, 3000–1650 cm.$^{-1}$, the only significant changes in spectrum during polymerization were the gradual disappearance of NCA carbonyl absorption (1850 and 1780 cm.$^{-1}$) and the appearance of absorption at 2330 cm.$^{-1}$ due to dissolved carbon dioxide. Polymerization proceeded at a negligible rate at −78°. When 0.2 equivalent of sodium methoxide was used as initiator at −78° there was an initial reaction (complete in 10 minutes) which destroyed NCA without producing the absorption expected for a carboxylic ester. After this reaction no further decrease in NCA absorption occurred so long as the reaction mixture was held at −78°. At about 0° polymerization proceeded at an appreciable rate.

CHICAGO, ILLINOIS

[CONTRIBUTION FROM THE DEPARTMENT OF BIOCHEMISTRY, UNIVERSITY OF KANSAS, AND THE WELLCOME RESEARCH LABORATORIES]

Oxygen Glycosides from the Hilbert–Johnson Pyrimidine Nucleoside Synthesis[1]

BY PHILIP NEWMARK AND IRVING GOODMAN

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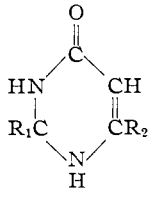
Synthesis of the nucleosides of 6-methyluracil (I), 6-methyl-2-thiouracil (II), 2-thiouracil (III) and barbituric acid (IV) was attempted by means of the Hilbert and Johnson pyrimidine nucleoside synthesis. No nucleosides were obtained. Both α- and β-isomers of the oxygen glucosides were isolated for I and II, and as one of the by-products, 4-ethoxy-6-methyl-2(1)-pyrimidone, all previously unreported. No condensation products were obtained for III or IV.

The classic reaction of Hilbert and Johnson,[2] involving the condensation of 2,4-dialkoxypyrimidines with acetylglycosyl bromides, was employed successfully by Hilbert and co-workers[3–5] and by others[6,7] for the preparation of uracil and cytosine nucleosides. Visser, Goodman and Dittmer[8] and Fox and Goodman[7] showed further that nucleosides of thymine and 5-methylcytosine could be obtained from the reaction of acetylglycosyl halides with 2,4-diethoxy-5-methylpyrimidines.

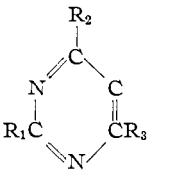
The work reported here concerns attempts by means of the Hilbert–Johnson reaction to synthesize nucleosides of several pyrimidines of biological interest which are not natural constituents of nucleic acids. We hoped in this way to prepare analogs of the naturally occurring nucleosides which might serve as metabolite antagonists for the normal nucleic acid derivatives. The pyrimidines selected were 6-methyluracil (I), 6-methyl-2-thiouracil (II), 2-thiouracil (III) and barbituric acid



	R_1	R_2
I	O	CH_3
II	S	CH_3
III	S	H
IV	O	OH

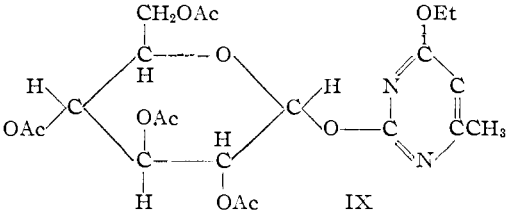
(IV). This study revealed some practical limitations of the reaction with respect to the structure of the pyrimidine moiety.

Condensation of tetra-O-acetyl-α-D-glucopyranosyl bromide with the ethoxy derivatives of 6-methyluracil and 6-methyl-2-thiouracil, V and VI respectively, yielded in each case both α- and β-

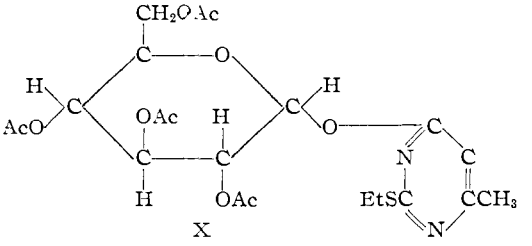


	R_1	R_2	R_3
V	OC_2H_5	OC_2H_5	CH_3
VI	SC_2H_5	OC_2H_5	CH_3
VII	SC_2H_5	OC_2H_5	H
VIII	OC_2H_5	OC_2H_5	OC_2H_5

isomers of the glucosides IX and X but no N-glucosylamines (nucleosides). Isolation of the α-form of IX from the same reaction was reported recently by Rabinowitz and Gurin.[9] Neither glucosides nor N-glucosylamines were obtained when the ethoxy derivatives of 2-thiouracil and barbituric acid, VII and VIII respectively, were allowed to react with tetra-O-acetyl-α-D-glucopyranosyl bromide. Although the use of the acetylglycosyl chlorides rather than the corresponding bromides increased the yields of the uracil and thymine nucleosides,[7] we obtained neither N-glycosylamines nor glycosides when we treated acetylglycosyl chlorides with the ethoxypyrimidines V,



IX



X

(1) This paper, which was presented in part at the 122nd Meeting of the American Chemical Society in Atlantic City, N. J., September, 1952, was the result of work initiated by the authors at the University of Colorado, with the aid of a fellowship (P.N.) and of a grant-in-aid (I.G.) from the U. S. Public Health Service. Nomenclature used in this article is in accord with the Rules of Carbohydrate Nomenclature, *Chem. Eng. News*, **31**, 1775 (1953).

(2) G. E. Hilbert and T. B. Johnson, THIS JOURNAL, **52**, 4489 (1930).

(3) G. E. Hilbert and E. F. Jansen, *ibid.*, **58**, 60 (1936).

(4) G. E. Hilbert, *ibid.*, **59**, 330 (1937).

(5) G. E. Hilbert and C. E. Rist, *J. Biol. Chem.*, **117**, 371 (1937).

(6) G. A. Howard, B. Lythgoe and A. R. Todd, *J. Chem. Soc.*, 1052 (1947).

(7) J. J. Fox and I. Goodman, THIS JOURNAL, **73**, 3256 (1951).

(8) D. W. Visser, I. Goodman and K. Dittmer, *ibid.*, **70**, 1926 (1948).

(9) J. L. Rabinowitz and S. Gurin, *ibid.*, **75**, 5758 (1953).