the combining region of the antibody a positive charge, or two positive charges, close to the negative charges of the two carboxylate ions of the hapten. The calculated distance between the negative charge of the carboxylate ion and the positive charge (assumed to be a single charge) in the antibody is 5.6 Å., which is 2.6 Å. greater than the minimum possible distance of approach of complementary charges in the two molecules.

A large decrease in combining power with the antibody is observed in the sequence phthalate ion, pyridine-2,3-dicarboxylate ion, pyrazine-2,3-dicarboxylate ion. This decrease is interpreted as resulting from hydration of the heterocyclic ions in solution. The energy required to break the hydrogen bonds between a heterocyclic nitrogen atom and an attached water molecule is calculated from the hapten-inhibition data to lie between 700 and 850 cal. mole^{-1}.

Unusually small steric effects of groups substituted in the phthalate ion have been observed. These observations are interpreted as showing that the antibodies fit only rather loosely about the haptenic group, the diametral dilatation being about 2 Å. in the region near the carboxyl ion groups and somewhat larger on the opposite side of the benzene ring. An explanation of the greater looseness of fit of these antibodies than of those in the systems previously studied is presented.

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[CONTRIBUTION FROM THE VENABLE CHEMICAL LABORATORY OF THE UNIVERSITY OF NORTH CAROLINA]

Reaction of Acetylglycine with Ethyl Orthoformate in Acetic Anhydride

BY RICHARD H. WILEY, OLIN H. BORUM AND LEONARD L. BENNETT, JR.[1]

The reaction of acetylglycine and ethyl orthoformate in acetic anhydride has been reported to be anomalous in that no 4-ethoxymethylene-5-oxazolone is obtained.[2] We wish to report the isolation of two new compounds from this reaction. One is diacetylglycine ethyl ester; the other a solid for which the formula $CH_3C(OC_2H_5){=}NCH_2CO{-}O{-}COCH_3$ is proposed. The same diacetyl ester has also been obtained in up to 88% yield from acetylglycine ethyl ester and acetic anhydride with or without pyridine. This ester reacts slowly with water and with butylamine to give acetylglycine N-*n*-butylamide, previously undescribed and N-*n*-butylacetamide. The ester is reduced with lithium aluminum hydride to diethylethanolamine.

$$(CH_3CO)_2NCH_2CO_2C_2H_5 \longrightarrow (C_2H_5)_2NHCH_2CH_2OH$$

The solid, m. p. 103–104°, obtained in the ethyl orthoformate reaction reacts readily with water to form acetylglycine quantitatively along with identifiable amounts of ethyl acetate; reacts with butylamine to form N-butylamides of acetylglycine and acetic acid; and is readily oxidized by permanganate. Analytical data show the addition of one acetyl and one ethyl group to acetylglycine. Of several possible structures, the ethoxyethylidene anhydride (A) and the N-ethyl anhydride (B) are considered likely although neither O- nor N- alkylation of amides by ethyl orthoformate has been established. The structure (A) is indicated by the reaction with water.

$$CH_3C(OC_2H_5){=}NCH_2CO{-}O{-}COCH_3$$

(A)

$$CH_3CON(C_2H_5)CH_2CO_2COCH_3$$

(B)

Hydrolysis of the imido ester to acetylglycine rather than glycine is possible by a reaction similar to the Schotten–Baumann reaction in which the mixed anhydride acetylates the amino group more rapidly than it is hydrolyzed by water.

Infrared absorption spectra[3] show an absorption band at 5.6 microns characteristic of the anhydride carbonyl; absence of the characteristic ester carbonyl absorption band at 5.8 microns and the presence of absorption bands at 6.0, 8.0, and 8.4 microns attributable to C=N, and C—O (C unsaturated) linkages by comparison with N-(ethoxyethylidene)-glycine ester. The absorption band at 5.6 microns, the only band the solid has in common with acetic anhydride, is also observed with phthalimide. This suggests a diacylamino structure for the solid which, however, is not consistent with other data. The spectra are given in Figs. 1 and 2.

Experimental

Reaction of Acetylglycine with Ethyl Orthoformate and Acetic Anhydride.—Acetylglycine, 50 g., and 260 g. of acetic anhydride were heated together with stirring for 20 min. at 90° to dissolve whereupon 63 g. of ethyl orthoformate was added. After heating at 90–100° for one hour and fractionating, 26–34 g. of product b.p. 126–136° at 4 mm. was collected. At 120–130°, 42–43 g. of this fraction was obtained. On standing 13–20 g. of solid separated from this liquid. The solid was separated by filtration, and the liquid purified by washing with potassium permanganate. Thus purified the product had b.p. 112° at 3.5 mm. and $n^{25}D$ 1.4532 and was similar in refractive index and analysis to the diacetylglycine ethyl ester isolated by fractionation from a refluxed mixture of acetylglycine ethyl ester and acetic anhydride with or without pyridine.

Anal. Calcd. for $C_8H_{13}O_4N$: C, 51.33; H, 7.00; N, 7.5; mol. wt., 187. Found: C, 51.27; H, 6.80; N,

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[1] Taken in part from the Ph.D. thesis of L. L. B.; address, Birmingham, Alabama.

[2] Barber, *et al.*, British Patent 585,089, January 30, 1947.

[3] The authors are indebted to Ralph M. Hill and Miss M. F. Bell, Esso Laboratories, Standard Oil Development Co., for generously providing these data.

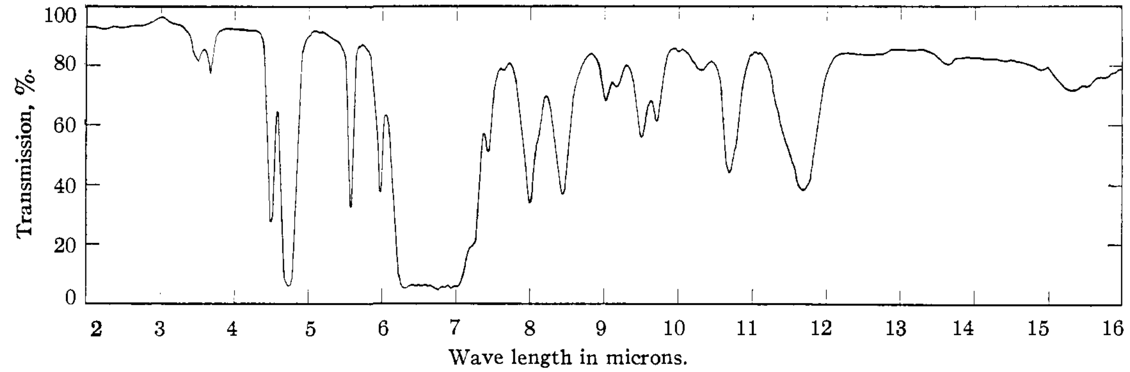

Fig. 1.—Infrared spectrum of solid, m. p. 103–104° in carbon bisulfide: absorption at 4.4–5.0, 6.2–7.2 and 11.2–12.0 microns is that of carbon disulfide.

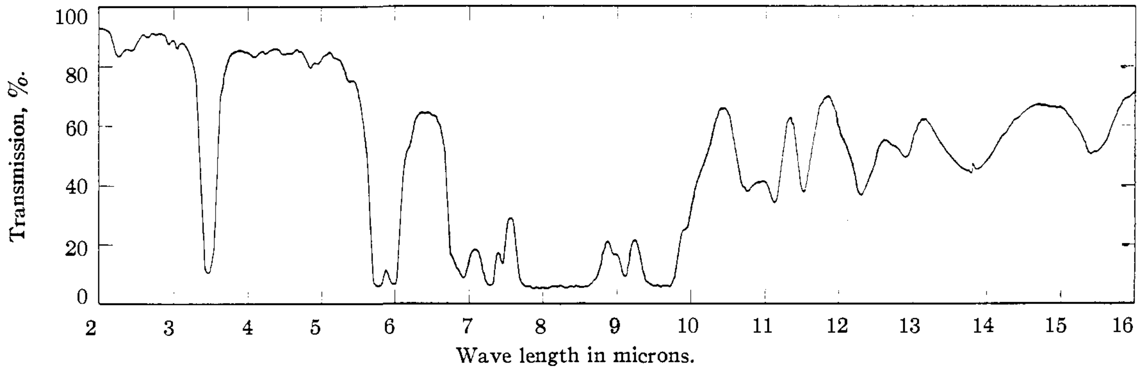

Fig. 2.—Undiluted ethoxyethylideneglycine ester

8.0; acetyl, 2.23/mol. wt., 187; ethoxyl, 1.07/mol. wt. 187.

Characterization of the Diacetyl Ester.—The diacetyl ester reacts with butylamine to give (1) N-butylacetamide, identified by comparison of refractive index, $n^{25}\mathrm{D}$ 1.4388, and b.p. with that of a synthetic sample, and (2) acetylglycine N-*n*-butylamide, m. p. 133–134° from nitromethane. The latter, previously unreported, was also prepared from acetylglycine ethyl ester, m.p. 134–134.5°, no depression of mixed m. p.

Anal. Calcd. for $C_8H_{16}O_2N$: C, 55.8; H, 9.4; N, 16.27. Found: C, 55.83; H, 8.84; N, 16.28.

The diacetyl ester, 9.35 g. (0.05 mole), dissolved in 50 ml. of anhydrous ether, was added over four hours to a solution of 5.68 g. (0.15 mole) of lithium aluminum hydride (Metal Hydrides Co.) in 100 ml. of anhydrous ether at 2–8°. After two hours at 2–25°, alcohol and then water were added to the mixture. The ether layer and ether extracts of the residue were combined, the ether removed, and the residue fractionated to give 2.50 g., 42.8% of diethylethanolamine, b.p. 159–161°, $n^{25}\mathrm{D}$ 1.4387–1.4390. The b.p. and n_D although in only fair agreement with values previously reported[4] agree precisely with values observed on a center cut of refractionated diethylethanolamine supplied by Carbide and Carbon Chemicals Corporation.[5]

Anal. Calcd. for $C_6H_{15}ON$: N, 11.95. Found: N, 12.18.

The diethylethanolamine was converted to its picrate, m. p. 77–77.5°; picrate of Carbide and Carbon sample, m. p. 78–79°; mixed m. p. 76–78°.

Anal. Calcd. for $C_{12}H_{18}O_8N_4$: N, 16.18. Found: N, 16.20.

Characterization of the Solid.—The solid was recrystallized from nitromethane, m. p. 103–104°.

Anal. Calcd. for $C_8H_{13}O_4N$: C, 51.33; H, 7.00; N, 7.50; mol. wt., 187. Found: C, 51.82; H, 7.07; N, 7.79; mol. wt., 174 ± 3 (ebull. in acetone); 177 ± 8 (ebull. in benzene); acetyl, 2.08/mol. wt. 187; ethoxyl 0.93/mol. wt. 187; Zerewitinoff active hydrogen, 0.

Reaction with water or butylamine gave products listed above, identified by combustion or neutral equivalent as well as mixed m. p. with synthesized samples. Reaction with butanol gave acetylglycine *n*-butyl ester, previously unreported, b. p. 139–141° (4 mm.).

Anal. Calcd. for $C_8H_{15}O_3N$: N, 8.09. Found: N, 8.10.

Summary

Acetylglycine reacts with acetic anhydride and ethyl orthoformate to give N,N-diacetylglycine ethyl ester, which is also formed from acetylglycine ethyl ester and acetic anhydride, and a solid m.p. 103–104° for which the formula $CH_3C(OC_2H_5){=}NCH_2COOCOCH_3$ is proposed.

N,N-Diacetylglycine ethyl ester is reduced by lithium aluminum hydride to diethylethanolamine.

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[4] Horne and Shriner, This Journal, 54, 2928 (1932).

[5] The authors appreciate the generous gift of this material.