

isobutyl-2-isopropylloxazolidine yielded besides the expected product some 2-diisobutylaminoethanol. This reducing reaction of some Grignard reagents has been observed by Conant<sup>8</sup> and by Kharasch<sup>9</sup> in their work on the reaction of Grignard reagents with some highly branched ketones. The dialkylamino alcohols prepared by this method are listed in Table III.

**Reaction of Grignard Reagents with 1-Aza-3,7-dioxabicyclo[3.3.0]octanes.**—The procedures for the reaction of Grignard reagents with 1-aza-3,7-dioxabicyclo[3.3.0]octanes and the isolation of products were the same as in the above reaction, except that the molar ratio of Grignard reagent to bicyclic compound was 2:1 in the experiments in which a 5-alkyl-1-aza-3,7-dioxabicyclo[3.3.0]octane or 1-aza-3,7-dioxabicyclo[3.3.0]octane was used and 3:1 in the experiment in which 5-hydroxymethyl-1-aza-3,7-dioxabicyclo[3.3.0]octane was used. The dialkylamino polyhydric alcohols that were prepared by this method are listed in Table IV.

### Discussion

The identity of the products from the hydrogenation of oxazolidines has been established by Cope and Hancock. The structures of the products from the reaction of Grignard reagents with N-substituted oxazolidines are supported by analytical data. Comparison of 2-diisobutylaminoethanol which was obtained by hydrogenating 3-isobutyl-2-isopropylloxazolidine with the product which was obtained by hydrolyzing the Grignard addition product from the reaction of isopropylmagnesium chloride with 3-isobutyl-oxazolidine reveals that the two materials are identical. Each product boiled at 89° at 10 mm., and the picrate of each product as well as a mixture of the two picrates melted at 122.5°. This additional datum adds further proof to the mechanism of the reaction of Grignard reagents with N-substituted oxazolidines.

The structures of the products from hydrogenation of 1-aza-3,7-dioxabicyclo[3.3.0]octanes as well as the structures of the products from the reaction of Grignard reagents with these bicyclic compounds are supported by analytical data.

(8) Conant and Blatt, *THIS JOURNAL*, **51**, 1227 (1929).

(9) Kharasch and Weinhouse, *J. Org. Chem.*, **1**, 209 (1936).

These data in turn support the proposed structures of the 1-aza-3,7-dioxabicyclo[3.3.0]octanes. The preparation of 2-dibutylamino-2-methyl-1,3-propanediol by the Grignard method (from *n*-propyl chloride and 5-methyl-1-aza-3,7-dioxabicyclo[3.3.0]octane) and by the hydrogenation method (hydrogenation of 2,8-dipropyl-5-methyl-1-aza-3,7-dioxabicyclo[3.3.0]octane) provides further proof of the structures of the 1-aza-3,7-dioxabicyclo[3.3.0]octanes and the mechanism of the reactions of these compounds which are proposed earlier in the paper and reported herein.

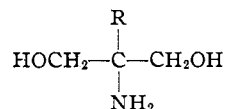
### Summary

Some new oxazolidines have been prepared from aldehydes and amino alcohols as well as N-alkylamino alcohols.

Some of the oxazolidines that were prepared were hydrogenated to N-alkylamino and N-dialkylamino alcohols.

The reaction of Grignard reagents with N-substituted oxazolidines was studied. A N-dialkylamino alcohol was isolated from each of these experiments. The mechanism for this reaction is discussed.

1-Aza-3,7-dioxabicyclo[3.3.0]octanes were prepared from some amino polyhydric alcohols of the type



and some aldehydes. Catalytic hydrogenation of these compounds gave dialkylamino polyhydric alcohols. The reaction of Grignard reagents with 1-aza-3,7-dioxabicyclo[3.3.0]octanes gave addition products which on hydrolysis yielded dialkylamino polyhydric alcohols. Mechanisms of these reactions are discussed.

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[CONTRIBUTION FROM THE BAKER LABORATORY OF CHEMISTRY AT CORNELL UNIVERSITY]

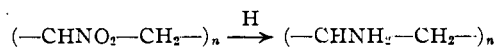
## Polymerization of Nitroolefins. The Preparation of 2-Nitropropene Polymer and of Derived Vinylamine Polymers

BY A. T. BLOMQUIST, W. J. TAPP AND JOHN R. JOHNSON

Despite the large variety of synthetic high polymers recorded in the chemical literature, there is no description of a polymeric amine with recurring primary amino groups attached directly to a long carbon chain, such as  $(-\text{CHNH}_2-\text{CH}_2-)_n$ . Polymers of acyl derivatives of vinylamine, such as N-vinylimide and vinyl isocyanate polymers, have been described but efforts to obtain a vinylamine polymer from these acyl derivatives or by other routes have failed.<sup>1</sup>

(1) Jones, Zomlefer and Hawkins, *J. Org. Chem.*, **9**, 500 (1944). This paper surveys various methods for obtaining vinylamine polymers.

The reported polymerization of nitroethylene<sup>2</sup> and 1-nitropropene<sup>3</sup> suggested a possible indirect synthesis of a vinylamine polymer by reduction of an appropriate nitroolefin polymer.

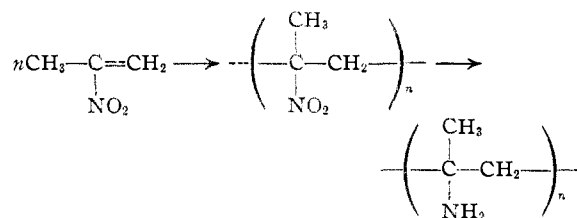


Attempts to reduce nitroethylene polymer to vinylamine polymer have not succeeded,<sup>1</sup> but this may be due to the circumstance that nitroethylene polymer is probably highly cross-linked

(2) Wieland and Sakellerios, *Ber.*, **52**, 898 (1919).

(3) Schmidt and Rutz, *ibid.*, **61**, 2142 (1928).

through reactions involving the active hydrogen of  $-\text{CH}(\text{NO}_2)-$  groupings. In order to avoid undesirable complications arising from such structures, the tertiary nitroolefin 2-nitropropene was chosen for the present study. Simple vinyl polymerization of this nitroolefin would afford a structure in which the nitro group is attached to a tertiary carbon atom. Subsequent reduction of such a polymer might be expected to give the related vinylamine polymer.



The simple 2-nitroolefin, 2-nitropropene, has not been described previously. The preparative methods used for 1-nitroolefins, such as dehydration of the appropriate nitroalcohol<sup>1</sup> and alkaline decomposition of the acetate derived from the nitroalcohol<sup>3,4,5,6</sup> gave poor yields when applied to 2-nitro-1-propanol. Inasmuch as olefins frequently may be obtained in good yield by pyrolysis of higher esters,<sup>7</sup> such as the benzoate or phenylacetate, experiments were made with the benzoate of the nitroalcohol. Pyrolysis of the benzoate of 2-nitro-1-propanol afforded 2-nitropropene (b.p. 48–49° (60 mm.)) in 54% yield. This nitroolefin is a yellow-green liquid which has strong lachrymatory properties and rapidly undergoes alteration on standing.

It was found that the polymerization of 2-nitropropene could be carried out readily by the method of Schmidt and Rutz<sup>3</sup> using aqueous potassium bicarbonate as a catalyst. The resulting 2-nitropropene polymer is a pale yellow, microcrystalline solid which decomposes without melting when heated above 200°. The nitropropene polymer is moderately soluble in organic solvents such as acetone, dioxane and acetic anhydride. Molecular weight determinations on this polymer have not as yet given reliable values but it appears likely that the molecular weight is relatively low.

Although a search of the literature failed to reveal reports of the polymerization of nitrobutenes<sup>8,9,10,11</sup> and higher nitroolefins, a few preliminary experiments in this laboratory indicate that a polymer can be obtained from 2-nitro-1-butene. An investigation of this nitroolefin and

its polymer is in progress and will be reported later.

2-Nitropropene polymer in dioxane or acetic anhydride solution was not reduced satisfactorily by metal-acid combinations or at low hydrogen pressures over Adams platinum catalyst but preliminary trials indicated that reduction could be accomplished by means of Adkins' technique at high pressures.<sup>12</sup> With Raney nickel catalyst 2-nitropropene polymer was smoothly hydrogenated in two hours at 90° under a pressure of 1700–1900 lb. using dioxane as a solvent. The resulting 2-aminopropene polymer is a reddish-brown glass which is soluble in water, dilute and concentrated acids, moderately soluble in dioxane and ethyl alcohol; it is insoluble in ether, acetone, hydrocarbons, and chlorinated hydrocarbons. Purification of the polymer was conveniently effected by conversion into either the phenylurea or phenylthiourea derivative. These derivatives are buff to yellow-brown microcrystalline solids that may be handled in the ordinary way.

The results of a series of catalytic hydrogenation

TABLE I  
HYDROGENATION OF 2-NITROPROPENE POLYMER

Run	Temp., °C.	Pressure, lb./sq. in.	Time, hr.	% Yield, crude amine	% Amine, insoluble in dioxane
1	135	1900	6	58.2	15.0 <sup>a</sup>
2	150	1850	8	59.4	53.4
3	150	1800	4	59.5	47.1
4	130	1800	3	59.5	2.6
5	110	1750	3	58.0	0.0
6	90	1750	3	64.1	0.0
7	90	1700	2	58.8	0.0

<sup>a</sup> Calcd. for  $\left( \begin{array}{c} \text{CH}_3 \\ | \\ -\text{C}-\text{CH}_2- \\ | \\ \text{NH}_2 \end{array} \right)_n$ : N, 24.53. Found: N, 16.04.

TABLE II  
PHENYLUREA DERIVATIVE FROM DIOXANE-SOLUBLE FRACTION

Run <sup>a</sup>	N, % <sup>b</sup>	Mol. wt. <sup>c</sup>	No. of monomeric units
1	14.96	2700	15
2	12.43	3200	18
3	12.62	2400	13
4	15.65	3700	21
5	15.45	4200	24
6	15.62 <sup>d</sup>	4200	24
7	15.59 <sup>d</sup>	5000	28

<sup>a</sup> Run numbers correspond to those given in Table I.

<sup>b</sup> Calcd. for  $\left( \begin{array}{c} \text{CH}_3 \\ | \\ -\text{C}-\text{CH}_2- \\ | \\ \text{NHCONHC}_6\text{H}_5 \end{array} \right)_n$ : N, 15.90. <sup>c</sup> Mean value of molecular weight ( $\approx 200$ ) determined by relative viscosity method of Staudinger. <sup>d</sup> See also Table III.

(12) We are indebted to Dr. H. R. Snyder of the University of Illinois for carrying out these preliminary high-pressure reduction studies.

(4) Hass, Hodge and Vanderbilt, *Ind. Eng. Chem.*, **28**, 339 (1936); **32**, 34 (1940).

(5) Schwarz and Nelles, U. S. Patent 2,257,980 (1941); *C. A.*, **36**, 494 (1942).

(6) Nightingale and Janes, *This Journal*, **66**, 352 (1944).

(7) Klages and Allendorf, *Ber.*, **31**, 1003 (1898); Hibbert and Burt, *This Journal*, **47**, 2240 (1925).

(8) Bouveault and Wahl, *Compt. rend.*, **131**, 1211 (1900).

(9) Bouveault and Wahl, *ibid.*, **134**, 1145 (1902).

(10) Haittinger, *Ann.*, **193**, 366 (1878).

(11) Haittinger, *Monatsh.*, **2**, 286 (1881).

experiments with 2-nitropropene polymer under a variety of conditions are presented in Table I. All of the hydrogenations described were carried out with 2-nitropropene polymer from a single preparation and all of the runs were made with nickel catalyst from the same preparation. The hydrogenation product was separated into a dioxane-soluble fraction and a dioxane-insoluble fraction. Data relating to composition and molecular weight of the dioxane-soluble fraction were obtained by examination of the phenylurea derivative formed by treatment of the polyamine with phenyl isocyanate (Table II).

From the data in Tables I and II it appears that hydrogenation of the nitroolefin polymer under a pressure of 1700–1750 lb. at 90–100° within a reaction time of two to three hours gives a product which is completely soluble in dioxane (runs 5, 6, and 7) and contains approximately 24–28 2-amino-propene units. Nitrogen analyses of the phenylurea derivative (N, 15.45–15.62%) are in good agreement with the value calculated for a phenylpropenylurea unit (calcd. for  $\begin{array}{c} \text{CH}_3 \\ | \\ \text{---C---CH}_2\text{---} \\ | \\ \text{NHCONHC}_6\text{H}_5 \end{array}$ , N, 15.90%). Analyses of two other derivatives, the phenylthiourea and *p*-bromophenylurea (Table III), also indicate a similar 2-amino-propene unit for the polyamine.

TABLE III<sup>a</sup>  
DERIVATIVES OF 2-AMINOPROPENE POLYMER

Run <sup>b</sup>	Derivative	Formula	Calcd. % N	Found
6	Phenylurea	(C <sub>11</sub> H <sub>12</sub> N <sub>2</sub> O) <sub>n</sub>	15.90	15.62
6	Phenylthiourea	(C <sub>10</sub> H <sub>12</sub> N <sub>2</sub> S) <sub>n</sub>	14.55	14.72 <sup>c</sup>
7	Phenylurea	(C <sub>10</sub> H <sub>12</sub> N <sub>2</sub> O) <sub>n</sub>	15.90	15.59
7	<i>p</i> -Bromophenylurea	(C <sub>10</sub> H <sub>11</sub> N <sub>2</sub> OBr) <sub>n</sub>	10.99	10.72 <sup>d</sup>
7	Phenylthiourea	(C <sub>10</sub> H <sub>12</sub> N <sub>2</sub> S) <sub>n</sub>	14.55	14.67 <sup>e</sup>

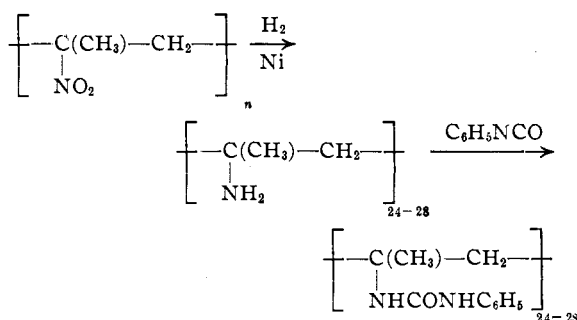
<sup>a</sup> Showing the analysis of different derivatives of 2-aminopropene polymer in comparison with those calculated for a regularly recurring 2-aminopropene unit. <sup>b</sup> Run numbers correspond to those given in Table I. <sup>c</sup> Calcd.: S, 16.67. Found: S, 16.52. <sup>d</sup> Calcd.: Br, 31.32. Found: Br, 30.26. <sup>e</sup> Calcd.: S, 16.67. Found: S, 16.18.

Hydrogenation of the 2-nitropropene polymer at higher temperatures and with longer reaction times (130–150° for four to eight hours) resulted in the formation of 15–53% of dioxane-insoluble polymer having a nitrogen content of 16%. Since the nitrogen content calculated for the aminopropene structural unit  $\begin{array}{c} \text{---C(CH}_3\text{)---CH}_2\text{---} \\ | \\ \text{NH}_2 \end{array}$  is

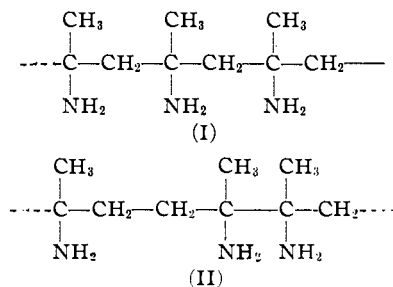
24.5%, a simple calculation shows that deamination occurred to the extent of about 35% in this material. The dioxane-soluble polymers formed under these conditions are of lower molecular weight and lower nitrogen content than the material formed under milder conditions. The phenylurea derivatives from the dioxane-soluble fractions show a molecular weight corresponding to 13–18 structural units instead of 24–28; the ni-

trogen content of 12.62–14.96% is appreciably less than that calculated for a phenylpropenylurea unit (N = 15.90%), and indicates that in this fraction the extent of deamination was only 6–20%. Ammonia was formed to some extent even under the mildest conditions of reduction, but in these runs the deamination was very slight (<3%) since the phenylurea derivatives had a nitrogen content in good agreement with that calculated for a phenylpropenylurea unit. As the same type of nitropropene polymer was used in all of the hydrogenations, it appears that the more drastic conditions bring about degradation of the polymer chain and also cross-linking of the chains.

The reactions involved in reduction of 2-nitropropene polymer under mild conditions and in characterization of the soluble polyamine are shown below. Analytical data for the derivatives

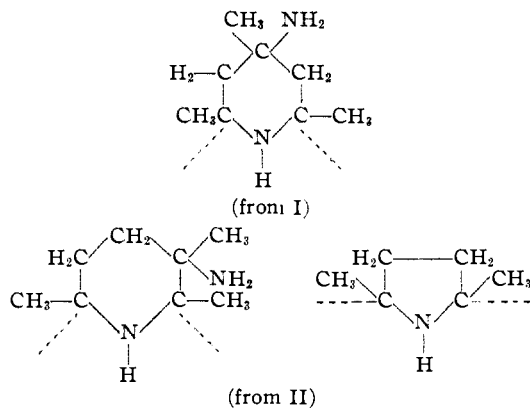


(Table III) are consistent with the formulation of the polyamine as a structure of recurring 2-aminopropylene units. No evidence is at hand to indicate whether the polymer is of the head-to-tail type (I) or head-to-head, tail-to-tail type (II). In the former all of the amino groups have a

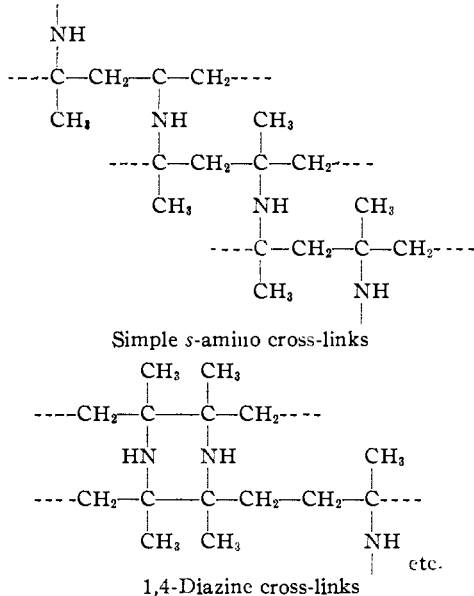


1,3-diamine and 1,3,5-triamine relationship, but the latter contains 1,2- and 1,4-diamine units and 1,2,5-triamine groups. The action of nitrous acid on an aqueous solution of the dioxane-soluble polyamine brought about evolution of gas and gave a brick-red, water-insoluble material which resisted purification and has not yet been characterized.

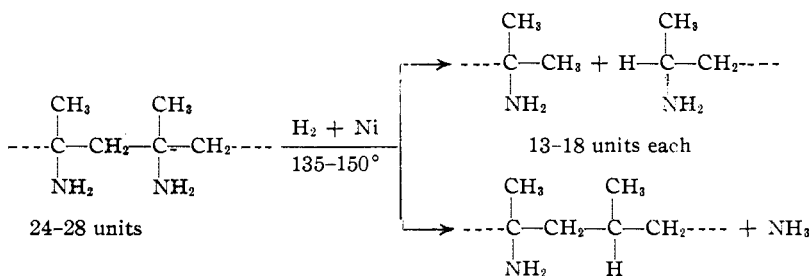
Inasmuch as ammonia was eliminated to a small extent during formation of the dioxane-soluble polyamine, it is possible that deamination occurred between amino groups in the 1,4- or 1,5-positions to form a pyrrolidine or aminopiperidine unit



None of the evidence at hand excludes secondary amino groups, but the amount of ammonia lost is less than that required for the formation of one of these rings in a chain of 24–28 units. On the other hand, the more drastic reductions leading to the formation of dioxane-insoluble polyamine may bring about intramolecular cyclizations and also effect cross-linking through various types of intermolecular deaminations:



The formation of a partially deaminated, dioxane-soluble fraction of lower molecular weight



(13–18 units) under the vigorous conditions of

hydrogenation indicates that hydrogenolysis of the carbon chain of the polyamine (or the nitroolefin polymer) has occurred to an appreciable extent, and possibly also hydrogenolysis of carbon–nitrogen bonds, as shown.

It is hoped that detailed studies of the dioxane-insoluble polymers now in progress will afford an insight into their constitution and mode of formation.

### Experimental

**Preparation of 2-Nitropropene.**—2-Nitro-1-propanol was prepared conveniently by the procedure of Vanderbilt and Hass,<sup>4</sup> and converted to 2-nitropropyl acetate using the general method described by Tindall.<sup>13</sup> The preparation of 2-nitropropyl benzoate is described below.

**2-Nitropropyl Benzoate.**—In a 2-liter round-bottomed flask equipped with a reflux condenser connected to a gas adsorption trap, a mixture of 189 g. (1.8 moles) of 2-nitropropanol, 222.6 g. (189 cc., 1.8 moles) of benzoyl chloride, and 450 cc. of benzene was heated to boiling. The reaction mixture was refluxed for twelve to fourteen hours. After cooling, the benzene was removed under reduced pressure (100 mm.). To the cooled liquid there was added 100 cc. of 95% ethyl alcohol, and the ethyl benzoate formed was removed by vacuum distillation at 30 mm. pressure and 100°. The reddish-brown residue was taken up in 500 cc. of ether and washed successively with two 100-cc. portions of water, three 100-cc. portions of saturated aqueous sodium bicarbonate solution, two 50-cc. portions of 10% hydrochloric acid, and three 100-cc. portions of water. The ether solution was dried over anhydrous sodium sulfate, with frequent shaking, for twelve to twenty-four hours. After removal of the ether under diminished pressure, 319.5 g. of the crude benzoate was obtained (87% yield).

Several attempts were made to purify the ester by distillation, but even at pressures of 1–1.5 mm. decomposition occurred and the only volatile products isolated were benzoic acid and 2-nitropropene.

**2-Nitropropene.**—In a 50-cc. Claisen flask attached to a water-cooled condenser was placed 21.0 g. (0.1 mole) of 2-nitropropyl benzoate, and the flask was heated in an oil-bath. Decomposition of the ester began at an oil-bath temperature of about 190–195° and the crude olefin distilled at 110–140°. After the distillation ceased, the deep blue-green distillate was taken up in 20 cc. of absolute ether and dried for six to eight hours over anhydrous copper sulfate.

The ether solutions of crude nitroolefin from nine 21-g. portions of 2-nitropropyl benzoate were combined and distilled. After removal of the ether the nitroolefin was obtained as a fraction boiling from 45–50° (60–62 mm.). Fractionation of this distillate through a heated 25-cm. column packed with glass helices gave a product boiling at 48–49° (59.5 mm.) (uncor.). There was obtained 42.0 g. (83.6% yield) of a yellow-green liquid which was highly lachrymatory and which became red-brown on standing:  $d_{20}^{20}$  1.0492;  $n_D^{20}$  1.4292;  $M_D$  calcd., 21.21; found, 21.2.

Pyrolysis of larger portions of 2-nitropropyl benzoate (35 to 45 g.) resulted in appreciably diminished yields. Combining the products from several 21-g. portions increased the yield over that obtained from single portions by decreasing the mechanical losses.

**Polymerization of 2-Nitropropene.**—In a 250-cc. wide-mouthed flask a mixture of 22.0 g. (0.253 mole) of freshly prepared 2-nitropropene (material boiling at 45–49° (60 mm.) was satisfactory) and 60 cc. of saturated aqueous

(13) Tindall, *Ind. Eng. Chem.*, **33**, 65 (1941).

potassium bicarbonate was stirred vigorously with a motor-driven stirrer. After a period of about fifteen minutes the mixture became milky in appearance and the temperature rose to 50°. Shortly after coagulation of the polymer occurred, giving a greenish-white paste, which stalled the stirrer. This pasty material, containing entrapped carbon dioxide, quickly hardened to a solid brittle mass. The solid was filtered off, ground with distilled water in a mortar and again filtered with suction, and finally washed with water until the filtrate was no longer alkaline. After air drying, the polymer weighed 20.5 g. (93.2% yield). The polymer decomposed without melting above 200° but the decomposition temperature varied widely with change in the rate of heating.

*Anal.* Calcd. for  $C_3H_5O_2N$ : C, 41.36; H, 5.79; N, 16.01. Found: C, 42.35; H, 5.62; N, 15.98.

**2-Aminopropene Polymer.**—Since several catalytic reductions of the nitropropene polymer have been carried out, varying only conditions of time, temperature and pressure, only one typical procedure is described.

In a copper bomb-liner, of about 450-cc. capacity, was placed a solution of 10 g. (0.115 mole) of 2-nitropropene polymer in 100 cc. of peroxide-free dioxane,<sup>14</sup> and 5 g. of Raney nickel catalyst.<sup>15</sup> The liner was enclosed in a steel bomb, similar to that described by Adkins<sup>16</sup> for hydrogenations at pressures of 100 atmospheres or more. Hydrogen was introduced from a commercial tank until a gage pressure of 1700 lb./sq. in. was attained. The contents of the bomb were rocked by means of a mechanical rocker equipped with an insulated heating element. The temperature of the contents of the bomb was maintained at 90 ± 2°. After the reaction had proceeded for two hours, the rocking and heating were stopped. When the bomb had cooled to room temperature the gage pressure was 1500 lb./sq. in. The approximate pressure drop as calculated by Adkins' empirical formula<sup>17</sup> was 100 lb./sq. in.

After the pressure had been released, the copper liner was removed from the bomb and opened. A strong odor of ammonia was noted. The contents of the bomb were carefully washed onto a Buchner funnel and the nickel was washed with four 10-cc. portions of dioxane. Evaporation of the pink colored dioxane solution to dryness gave a reddish brown glass; yield 4.5 g. (68.7%). The decomposition temperature of the polymer varied between 145 and 165° depending upon small variations in the rate of heating.

Aqueous solutions of the polymeric amine when treated with nitrous acid gave a brick-red water-insoluble tar together with copious evolution of gas. The tar was not investigated.

#### Derivatives of 2-Aminopropene Polymer

**N-Phenylurea Derivative.**—In a 200-cc. flask was placed 7.8 g. (0.137 mole) of 2-aminopropene polymer dissolved in 100 cc. of dioxane. To this was added 16.8 g. (15.2 cc., 0.141 mole) of phenyl isocyanate in dropwise fashion, with vigorous stirring. The mixture, which became dark brown and appreciably warmer, was heated on a steam-bath for two hours. On cooling, the reaction mixture was poured into 500 cc. of ether with stirring. The phenylurea derivative separated immediately as a buff-colored microcrystalline precipitate. The product was filtered and washed successively with six 25-cc. portions of ether, three 10-cc. portions of water, six 25-cc. portions of ether and dried in air. It softened between 135 and 150° and decomposed between 150 and 158° (uncor.), dependent upon the rate of heating; yield, 17.5 g. (73.3%).

*Anal.*<sup>17</sup> Calcd. for  $C_{10}H_{12}N_2O$ : C, 68.53; H, 6.33; N,

(14) Fieser, "Experiments in Organic Chemistry," 2nd ed., D. C. Heath and Company, New York, N. Y., 1941, pp. 368-369.

(15) Drake, "Organic Syntheses," Vol. 21, John Wiley and Sons, New York, N. Y., 1941, p. 15.

(16) Adkins, "Reactions of Hydrogen," University of Wisconsin Press, Madison, Wis., 1937.

(17) Analytical and molecular weight data for other phenylurea derivative preparations are given in Table I.

15.90. Found: C, 68.35; H, 6.48; N, 15.65; mol. wt., 3190.

The molecular weight value for the sample described above was determined by observing the freezing point depression in purified ethylene dibromide using the method of Reilly and Rae.<sup>18</sup>

Using this sample as a standard, relative viscosities of the phenylurea derivatives listed in Table I were measured. The measurements were carried out at 30.0 ± 0.1° using 0.500 g. samples in 10.0 cc. of reagent-grade acetone. The molal concentration of the samples was made the same and the value for the Staudinger constant ( $K$ ) should be the same for the different samples. The following relationship was used

$$\frac{N_{sp}}{N'_{sp}} = \frac{c \times K \times 3200}{c' \times K' \times M'}$$

where  $N_{sp}$  and  $N'_{sp}$  are the specific viscosities for the standard and the given sample, respectively, and  $M'$  is the molecular weight of the given sample. Since  $c$  and  $c'$  are equal, and  $K$  and  $K'$  should be the same, the proportion becomes

$$\frac{N_{sp}}{N'_{sp}} = \frac{3200}{M'}$$

It is realized that the Staudinger equation is not highly accurate and that the molecular weights determined by this method must be assumed to be approximations of the actual values. This method does, however, lend itself conveniently to observing a trend in average molecular weights for a series of comparable preparations.

**N-Phenylthiourea Derivative.**—This derivative was obtained using phenyl isothiocyanate, with the same conditions for preparation, isolation and purification described above. From 2.0 g. of 2-aminopropene polymer there was obtained 3.4 g. (50.6% yield) of the phenylthiourea derivative as a yellow-brown microcrystalline solid. Upon heating, it decomposed between 139 and 148° depending upon the rate of heating.

*Anal.* Calcd. for  $C_{10}H_{11}N_2S$ : N, 14.55; S, 16.67. Found: N, 14.72; S, 16.52

**N-*p*-Bromophenylurea Derivative.**—To a solution of 5.3 g. (0.027 mole) of *p*-bromophenyl isocyanate in 100 cc. of hot dioxane was added with stirring a solution of 1.5 g. (0.026 mole) of 2-aminopropene polymer in 15 cc. of dioxane. The reaction mixture was heated for two hours on the steam-bath. After cooling, it was poured with stirring into 300 cc. of ether. The *p*-bromophenylurea derivative separated as a tan colored microcrystalline precipitate which was washed successively with several 10-cc. portions of ether, twice with 10-cc. portions of water, and again with several 10-cc. portions of ether. The product was dried in air; yield 3.9 g. (58.9% of the theoretical).

*Anal.* Calcd. for  $C_{10}H_{11}N_2OBr$ : N, 10.99; Br, 31.32. Found: N, 10.72; Br, 30.26.

#### Summary

2-Nitropropene undergoes a smooth and rapid polymerization in the presence of aqueous potassium bicarbonate solution. Catalytic hydrogenation of 2-nitropropene polymer in dioxane with Raney nickel under mild conditions affords a water-soluble polymer which appears to be a linear polymer with primary amine groups, composed of 24-28 recurring 2-aminopropene units.

Hydrogenation of the nitroolefin polymer under more drastic conditions is accompanied by extensive deamination and yields a lower molecular

(18) Reilly and Rae, "Physico-chemical Methods," 2nd rev. ed., Methuen and Co., London, 1933, pp. 430-440.

weight dioxane-soluble polyamine as well as a dioxane-insoluble product. The dioxane-soluble polyamines have been characterized by the forma-

tion of phenylurea, phenylthiourea and *p*-bromo-phenylurea derivatives.

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## The Empirical Formula of $\beta$ -Lactoglobulin<sup>1</sup>

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From the point of view of organic chemistry, the establishment of the empirical formula of proteins (*cf.* Mulder<sup>2</sup> in 1838), in terms of constituent amino acids, is fundamental to an understanding of their constitution and to the development of theories of protein structure.

An apparently complete analysis of a simple, crystalline, homogeneous protein of small molecular weight (42,000), *viz.*,  $\beta$ -lactoglobulin<sup>3</sup> is presented in this paper. This protein, which constitutes the greater part of the whey proteins, was isolated in crystalline form by Palmer<sup>3,4</sup> from the plasma of cow's milk. It is free from phosphorus and carbohydrate. Physico-chemical studies<sup>5</sup> indicate that  $\beta$ -lactoglobulin is a protein native to milk and that it is homogeneous as tested by the solubility method,<sup>3</sup> in the ultracentrifuge,<sup>5</sup> by electrophoresis<sup>5</sup> and with respect to dissociation.<sup>6</sup>

The results are given in Table I; the analytical values for the constituents are given in % in column 4, in % amino acid residue in column 5 and in atoms or moles per 10<sup>5</sup> g. of protein in column 7. Columns 6 and 8-14 in Table I are based upon the minimum molecular weight, the calculation of which is shown in Table III. The data will first be discussed without any reference to the molecular weight and subsequently on a molar basis.

The following assumptions have been made:

(1) On the basis of physico-chemical criteria quoted above, it is assumed that  $\beta$ -lactoglobulin is a chemical compound in which the constituent amino acids are present in integral molar quantities.

(2) It is assumed that the amino acids (RNH<sub>2</sub>-COOH) are present in the molecule in peptide linkage. The yield is, therefore, given in per cent. of amino acid in column 4 and in per cent. of amino acid residue [R(NH—)(CO—)] in columns 5 and 6.

(3) Part of this work was carried out under contract, recommended by the Committee on Medical Research, between the Office of Scientific Research and Development and Columbia University. Much of the present data was obtained in the course of the development and standardization of methods for application to plasma proteins.

(2) Mulder, *Ann.*, **28**, 73 (1838).

(3) Palmer, *J. Biol. Chem.*, **104**, 359 (1934).

(4) *Cf.* Sørensen and Sørensen, *Compt. rend. trav. lab. Carlsberg*, Series C, **23**, 55 (1938).

(5) Pedersen, *Biochem. J.*, **30**, 948, 961 (1936).

(6) Sørensen and Palmer, *Compt. rend. trav. lab. Carlsberg*, Series C, **21**, 283 (1938).

(3) It is assumed that the amide nitrogen (detrn. 8) or amide NH<sub>2</sub> (detrn. 8a) as obtained by our method indicates the amount of glutamine and asparagine present. Since it is not yet possible to differentiate between these two amides, all of the amide NH<sub>2</sub> has been arbitrarily assigned to glutamine in detrn. 8b, and will subsequently be discussed as glutamine only. Consequently, all of the aspartic acid is considered as "free aspartic acid," while the "free glutamic acid" (detrn. 30) is the difference between total glutamic acid and glutamine.

(4) In the interpretation of the data on the free amino nitrogen of the intact protein obtained in the manometric Van Slyke apparatus under our conditions<sup>7</sup> it is assumed (a) that no splitting with the liberation of reactive amino N occurs during the determination; (b) that all of the  $\epsilon$ -amino N of lysine is free and reactive; (c) that all of the terminal free  $\alpha$ -amino N of the peptide chain, or chains (detrn. 12), is free and reactive and, therefore, can be obtained by deducting the  $\epsilon$ -amino nitrogen (calculated from the lysine value (detrn. 27a)) from the total free amino N (detrn. 11); (d) that no proline or hydroxyproline is at the end of a peptide chain; (e) that one free terminal COOH group corresponds on the other end of the chain to each terminal  $\alpha$ -amino group. In the accounting of the split products in per cent. of amino acid residue (detrn. 12a) in columns 5 and 6, a value for "Terminal H<sub>2</sub>O" corresponding to the terminal  $\alpha$ -amino N has, therefore, been added (*i. e.*, one H atom in terminal  $\alpha$  NH<sub>2</sub> and one OH group in terminal carboxyl).

(5) It is assumed that our determinations refer to the unhydrated molecule, since they were carried out on air dried material, corrected for moisture (*cf.* refs. 8, 9). We have previously contributed some evidence in favor of this contention in experiments in which H<sub>2</sub>O containing the heavy oxygen isotope O<sup>18</sup> was used as an indicator (*cf.* ref. 9, p. 374).

Two preparations of  $\beta$ -lactoglobulin were used for the experiments, for one we are indebted to Dr. R. K. Cannon, for the other to Dr. G. Haugaard. About 2.5 g. of protein was used for the determinations reported in Table I.

(7) Brand and Diskant, unpublished work.

(8) Brand and Kassel, *J. Gen. Physiol.*, **25**, 167 (1941).

(9) Brand and Kassel, *J. Biol. Chem.*, **145**, 365 (1942).