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Theoretical Analysis of the Polymerization Kinetics of N-Carboxy-α-amino Acid Anhydrides. II. An Amine Initiated Polymerization with a Two Stage Propagation[1]

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A theoretical analysis is given for the kinetics of a primary amine-initiated polymerization of N-carboxy-α-amino acid anhydrides in which the propagation reaction exhibits two successive rates depending on whether the degree of polymerization j of the growing peptide chains is smaller, or equal to or larger, than a given value s. In the calculations made it was assumed that no termination reaction takes place and that the initiation reaction is fast in comparison with the propagation reaction. Formulas are derived for the rate of polymerization and the molecular weight distribution of the polymer formed. A method is suggested for a rigorous derivation of the transition chain length s from experimental data. Values of $s = 8$ and 13 are derived for the polymerization in dioxane of N-carboxy-γ-benzyl-L-glutamate anhydride and N-carboxy-O-carbobenzoxy-L-tyrosine anhydride, respectively. Means are given for estimation of the number and weight fraction, as well as the number and weight average degrees of polymerization, of both the β and the α-fractions: the former composed of peptides with $j > s$, and the latter of peptides with $j \geq s$.

In a previous communication[1] a theoretical analysis of a primary or secondary amine initiated polymerization of N-carboxy-α-amino acid anhydrides was given. Equations for the kinetics of polymerization as well as for the expected molecular weight distribution were derived for a general case in which the polymerization consists of an initiation, a propagation and a termination reaction. Each of the individual reactions was assumed to be characterized by a single specific rate constant. Recent data[3-7] indicate, however, that when the amine initiated polymerization of N-carboxyamino acid anhydrides is carried out in solvents such as dioxane, benzene, chloroform or ethylene dichloride, the propagation reaction may be divided into two separate stages, each characterized by its own specific rate constant. For example, when N-carboxy-γ-benzyl-L-glutamate anhydride was polymerized in dioxane using n-hexylamine as initiator, the rate constant for the second propagation step was at least five times greater than that for the first.[3] Furthermore, the molecular weight distribution of the polymer obtained was much broader than could be explained on the basis of a single propagation rate constant. It was suggested[3] that the change in the rates of propagation is associated with a configurational change of the growing polymer chains, since the transition from the slow to the fast propagation rate occurred at an average chain length corresponding to 7 to 13, *i.e.*, the region assigned to the critical length for stabilization of the α-helical configuration. This suggestion has been corroborated by the findings[4,8] that polypeptide chains with degrees of polymerization smaller than those encountered in the transition region exist in dioxane solution in the β-form, while those of higher chain length are in the α-helical form.

A two stage propagation polymerization also has been observed (unpublished observations of J. D. Coombes, E. Katchalski and P. Doty) while studying the n-hexylamine initiated polymerization of N-carboxy-O-carbobenzoxy-L-tyrosine anhydride in dioxane (see Fig. 1). Ballard and Bamford[7] likewise have reported a two stage propagation polymerization for various N-carboxyamino acid anhydrides when initiated by primary amines in different organic solvents. However, these authors attribute the increase in polymerization rate to the formation of an insoluble polypeptide fraction which preferentially adsorbs the N-carboxyanhydride monomer.

A quantitative theoretical analysis of the above types of polymerization seems desirable, since low molecular weight peptides differ from those of high molecular weight, both in their conformation and physicochemical properties. The characterization of a given poly-α-amino acid sample, prepared under conditions where a two stage propagation reaction occurred, requires, therefore, a knowledge of the molecular weight distribution and the relative amounts of the polymer in the α and β forms.

The treatment to be described below deals with a primary amine initiated polymerization of N-carboxyamino acid anhydrides in which no termination reaction takes place and the initiation reaction is fast in comparison with the propagation reaction. The propagation reaction itself consists of two successive rates depending on whether the degree of polymerization of the growing peptide chains is smaller or equal to or greater than an initial value s. This treatment may be extended to a more complicated type of polymerization which includes a termination as well as a relatively slow initiation reaction. The mechanism of polymerization studied was chosen because of its relative simplicity and its resemblance to the type of polymerization found experimentally by Doty and Lundberg.[3] Finally it should be emphasized that the analytical procedure adopted is quite general and independent of the detailed mechanism responsible for the two stage propagation reaction.

Kinetic Equations.—In accord with the above it will be assumed that the two stage propagation polymerization may be described by the set of

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[1] For paper I dealing with amine initiated polymerization of N-carboxy-α-amino acid anhydrides see E. Katchalski, Y. Shalitin and M. Gehatia, This Journal, **77**, 1925 (1955).

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[3] R. D. Lundberg and P. Doty, This Journal, **79**, 3961 (1957); P. Doty and R. D. Lundberg, *ibid.*, **78**, 4810 (1956).

[4] M. Idelson and E. R. Blout, *ibid.*, **79**, 3948 (1957).

[5] H. Weingarten, *ibid.*, **80**, 352 (1958).

[6] W. J. Ritschard, *Makromolekulare Chem.*, **29**, 141 (1959); R. Buyle and B. Hargitay, Symposium über Makromoleküle in Wiesbaden, 1959, Verlag Chemie, GmbH. Weinheim/Bergstr.

[7] D. G. H. Ballard and C. H. Bamford, *J. Chem. Soc.*, 1039 (1959).

[8] M. Goodman and E. E. Schmitt, This Journal, **81**, 5507 (1959).

reactions

$$\mathrm{I} + \mathrm{M} \xrightarrow{k_0} \mathrm{M}_1 \qquad \text{(a)}$$

$$\mathrm{M}_j + \mathrm{M} \xrightarrow{k_1} \mathrm{M}_{j+1} \quad \text{for } j < s \qquad \text{(b)}$$

$$\mathrm{M}_j + \mathrm{M} \xrightarrow{k_2} \mathrm{M}_{j+1} \quad \text{for } j \geqslant s \qquad \text{(c)}$$

where I, M and M_j, denote the initiator, the reacting monomer and peptide chains composed of j amino acid residues, respectively. The propagation reaction of chains with a degree of polymerization j, where $j < s$, is characterized by a specific rate constant k_1, whereas for chains with $j \geqslant s$ the specific rate constant is k_2. The rate of reaction of monomer with initiator is assumed to be rapid compared to the rate of propagation, *i.e.*, $k_0 >> k_1$ or k_2. This assumption is in agreement with the experimental findings of Doty and Lundberg[3] and of Idelson and Blout.[4]

Chains composed of one amino acid residue disappear from the reaction mixture at a rate governed by eq. 1, where N_1 denotes the concentration of such chains and $\nu = \int_0^t (\mathrm{M})\mathrm{d}t$, so that $\mathrm{d}\nu = (\mathrm{M})\mathrm{d}t$. The rate of formation of chains composed of j amino acid residues is given by eq. 2 for $j = 1,2,\ldots, s - 1$, N_j denoting the concentration of the j-mers. Equation 3 describes the case for which $j = s$, and eq. 4 that in which $j = s + 1,\ s + 2,\ldots, \infty$.

$$\frac{\mathrm{d}N_1}{\mathrm{d}\nu} = -k_1N_1 \qquad (1)$$

$$\frac{\mathrm{d}N_j}{\mathrm{d}\nu} = k_1N_{j-1} - k_1N_j \qquad (2)$$

$$j = 1,2 \ldots (s-1)$$

$$\frac{\mathrm{d}N_s}{\mathrm{d}\nu} = k_1N_{s-1} - k_2N_s \qquad (3)$$

$$\frac{\mathrm{d}N_j}{\mathrm{d}\nu} = k_2N_{j-1} - k_2N_j \qquad (4)$$

$$j = s+1,\ s+2,\ \ldots, \infty$$

Upon integration, eq. 1 gives

$$N_1 = I_0e^{-k_1\nu} \qquad (5)$$

where I_0 denotes the concentration of initiator at $t = 0$, *i.e.*, at $\nu = 0$.

The successive solution of the set of equations 2, 3 and 4, gives

$$N_j = I_0\frac{(k_1\nu)^{j-1}}{(j-1)!}e^{-k_1\nu} \qquad (6)$$

$$j = 1,2,\ \ldots\ldots, s-1$$

$$N_s = I_0\frac{k_1^{s-1}}{(s-2)!}e^{-k_2\nu}\int_0^\nu \nu^{s-2}e^{\delta\nu}\,\mathrm{d}\nu \qquad (7)$$

where $\delta = k_2 - k_1$

$$N_j = I_0k_2^{j-s}\frac{k_1^{s-1}}{(s-2)!}e^{-k_2\nu}\underbrace{\int_0^\nu\cdots\int_0^\nu}_{\substack{j-s+1\\ \text{times}}} \nu^{s-2}e^{\delta\nu}\,\mathrm{d}\nu\ldots\mathrm{d}\nu \qquad (8)$$

$$j = s,\ s+1,\ s+2,\ \ldots\ldots, \infty$$

Equation 8 may be transformed readily into (9) as shown in Appendix I.

$$N_j = I_0k_2^{j-s}\frac{k_1^{s-1}}{(s-2)!}e^{-k_2\nu}\int_0^\nu \frac{y^{j-s}}{(j-s)!}e^{\delta(\nu-y)}(\nu-y)^{s-2}\,\mathrm{d}y \qquad (9)$$

$$j = s,\ s+1,\ s+2,\ \ldots\ldots, \infty$$

When $j = s$ eq. 9 reduces to eq. 7.

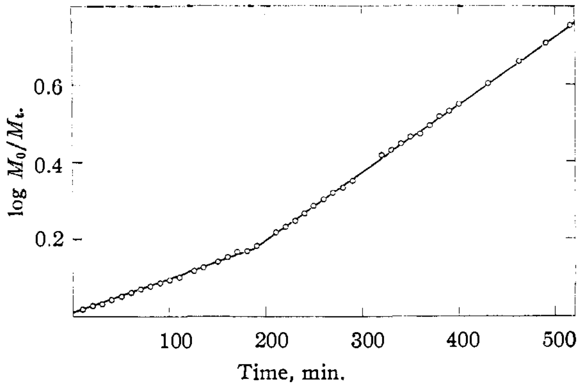

Fig. 1.—*n*-Hexylamine initiated polymerization of N-carboxy-O-carbobenzoxy-L-tyrosine anhydride in dioxane at 24.6°. Initial concentration of anhydride M_0 = 0.1173 mole liter^{-1}; that of initiator I_0 = 0.00293 mole liter^{-1}; the specific propagation rate constants k_1 = 0.707 liter mole^{-1} min.$^{-1}$ and k_2 = 1.36 liter mole^{-1} min.$^{-1}$ were obtained from the corresponding slopes of the curve.

For distribution calculations by means of a computer eq. 9 can be rewritten as a finite series (9a).

$$N_j = I_0k_2^{j-s}\frac{k_1^{s-1}}{(s-2)!}e^{-k_2\nu}\left\{\frac{\nu^{j-2}e^{\delta\nu}}{(j-s)!\delta}\sum_{r=0}^{j-s}\sum_{p=0}^{s-2+r}(-1)^{r+p}\binom{j-s}{r}\frac{(s-2+r)!}{(s-2+r-p)!(\delta\nu)^p} + \frac{\nu^{j-s}}{(j-s)!\delta^{s-1}}\sum_{r=0}^{j-s}(-1)^{s-1}\binom{j-s}{r}\frac{(s-2+\nu)!}{(\delta\nu)^r}\right\} \qquad \text{(9a)}$$

Equation 9a was derived from (9) by introducing the new variable $\nu - y = \nu x$ and solving for the integral $\int_0^1 e^{\delta\nu x}\ x^{s-2+r}\ \mathrm{d}x$[9a] after the expansion of the binomial $(1-x)^{j-s}$.

Equations 6, 7 and 9 give the concentrations of the growing peptide chains as a function of ν, and of the two propagation constants k_1 and k_2. The chain length distribution of the shorter chains with $j = 1,2,\ldots,s - 1$, is Poissonian in character and is encountered in the ethylene oxide type of polymerization[9b] when no termination occurs. The chain length distribution of the higher peptides with $j = s,\ s+1,\ \ldots, \infty$ is non-Poissonian and is governed by the two rate constants k_1 and k_2. However, when $k_2 = k_1$, equations 7 and 9 give (6) and the total distribution of the polymer becomes Poissonian.

The polymerization reaction under discussion is devoid of a termination reaction. The concentration of growing chains therefore remains constant throughout the reaction and equal to the initial concentration of initiator I_0. Hence

$$\sum_{j=1}^{\infty} N_j = I_0 \qquad (10)$$

Equation 10 may be verified by substitution of the appropriate values of N_j given by (6), (7) and (9).

Rate of Polymerization.—The rate of monomer consumption during the polymerization is given by

$$-\frac{\mathrm{d}M}{\mathrm{d}\nu} = k_1\sum_{j=1}^{s-1}N_j + k_2\sum_{j=s}^{\infty}N_j \qquad (11)$$

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(9) (a) W. Gröbner and N. Hofreiter, "Integraltafel," Part 2, Springer, 1950, p. 59. (b) P. J. Flory, THIS JOURNAL, **62**, 1561 (1940).

Substituting the values of N_j, $j = 1,2,\ldots, s-1$, given by eq. 6 and remembering that

$$\sum_{j=1}^{s-1} N_j + \sum_{s}^{\infty} N_j = I_0$$

one obtains

$$-\frac{\mathrm{d}M}{\mathrm{d}\nu} = k_2 I_0 - \delta I_0 e^{-k_1\nu} \sum_{j=1}^{s-1} \frac{(k_1\nu)^{j-1}}{(j-1)!} \qquad (12)$$

This equation indicates that at the beginning of the reaction when $\nu \rightarrow 0$

$$-\frac{\mathrm{d}M}{\mathrm{d}\nu} = k_1 I_0 \text{ or } -\frac{\mathrm{d}M}{\mathrm{d}t} = k_1 I_0 M$$

At this stage the polymerization reaction is first order with respect to monomer and governed by the rate constant k_1. Toward the end of the reaction, under conditions where it is justified to assume that $\nu \rightarrow \infty$

$$-\frac{\mathrm{d}M}{\mathrm{d}\nu} = k_2 I_0 \text{ or } -\frac{\mathrm{d}M}{\mathrm{d}t} = k_2 I_0 M$$

The reaction is again first order with respect to monomer but this time is characterized by the specific rate constant k_2. These conclusions are in accord with the experimental findings described above.

Evaluation of *s*.—The correlations developed suggest a suitable method for determining the chain length s, at which the transition in the specific rate of propagation occurs.

The derivative of (12) with respect to ν is

$$\frac{\mathrm{d}^2M}{\mathrm{d}\nu^2} = -\delta I_0 k_1 e^{-k_1\nu} \frac{(k_1\nu)^{s-2}}{(s-2)!} \qquad (13)$$

Since $\frac{\mathrm{d}^2M}{\mathrm{d}\nu^2} = 0$ only when $\nu = 0$ or $\nu = \infty$ (assuming $k_2 \neq k_1$), $\mathrm{d}M/\mathrm{d}\nu$ represents a monotonic function of ν. The third derivative given by

$$\frac{\mathrm{d}^3M}{\mathrm{d}\nu^3} = \delta I_0 k_1^2 e^{-k_1\nu} \frac{(k_1\nu)^{s-3}}{(s-3)!} \left[\frac{k_1\nu}{s-2} - 1\right] \qquad (14)$$

equals zero when

$$k_1\nu = s - 2 \qquad (14a)$$

The experimental determination of the value of ν (to be denoted as $\nu_{\text{transition}}$) at which $\frac{\mathrm{d}^3M}{\mathrm{d}\nu^3} = 0$ thus permits the evaluation of s.

When the transition in the rate of polymerization occurs at low monomer conversion $\left(s << \frac{M_0}{I_0}\right)$, then $\nu_{\text{trans.}} \simeq M_0 t_{\text{trans.}}$, and therefore

$$t_{\text{trans.}} = \frac{s-2}{k_1 M_0} \qquad (15)$$

Equation 15 shows that the time at which the transition occurs is independent of the concentration of initiator I_0 but is inversely proportional to the initial concentration of monomer. This may be expected since I_0 determines the number of growing chains, and not their rate of growth, while M_0 determines the rate of growth of any given chain.

The Fraction and Average Degree of Polymerization of Chains in the β-Form.—The total number of growing chains is given at any instant by I_0. The number of chains in the β-form (*i.e.*, $j < s$) is $\sum_{j=1}^{s-1} N_j$. Hence the fraction of chains in the β-form, P_n^β, is given by

$$P_n^\beta = \frac{1}{I_0} \sum_{j=1}^{s-1} N_j = e^{-k_1\nu} \sum_{j=1}^{s-1} \frac{(k_1\nu)^{j-1}}{(j-1)!} \qquad (16)$$

where we have made use of eq. 6.

In Fig. 2, P_n^β is presented as a function of $k_1\nu$ for various values of s. The figure shows that during the polymerization, as $k_1\nu$ increases, the fraction of chains in the β-form decreases gradually for any given value of s. Furthermore, if $k_1\nu$ is kept constant, an increase in s leads to a marked increase in the corresponding value of P_n^β.

The polymer weight fraction of chains in the β-form, P_w^β, is defined by

$$P_w^\beta = \frac{\sum_{j=1}^{s-1} jN_j}{\sum_{j=1}^{\infty} jN_j} \qquad (17)$$

Since $\sum_{j=1}^{\infty} jN_j = M_0 - M_t$, where M_0 denotes the initial concentration of monomer and M_t its concentration at time t, eq. 17 may be rewritten in the form

$$P_w^\beta = \frac{1}{M_0 - M_t} \sum_{j=1}^{s-1} jN_j \qquad (17a)$$

Inserting (6) into (17a) one obtains

$$P_w^\beta = \frac{I_0}{M_0 - M_t} e^{-k_1\nu} \sum_{j=1}^{s-1} \frac{j(k_1\nu)^{j-1}}{(j-1)!} \qquad (18)$$

A plot of $P_w^\beta \left(\frac{M_0 - M_t}{I_0}\right)$ *versus* $k_1\nu$, for different values of s, is given in Fig. 3. Since $P_w^\beta \times \left(\frac{M_0 - M_t}{I_0}\right)$ is proportional to the weight concentration of the β-fraction in the reaction mixture, the curves given illustrate the variation in the concentration by weight of the β-fraction with $k_1\nu$ and s. For any given value of s, the weight concentration of the β-fraction increases at the initial stages of the polymerization. However, as the reaction proceeds, the β-chains are transformed into α-chains and the weight concentration of the β-fraction decreases. The maximum amount of the β-fraction to be expected in any given experiment increases with s.

The number average degree of polymerization of the β-fraction is given by

$$DP_n^\beta = \frac{\sum_{j=1}^{s-1} jN_j}{\sum_{j=1}^{s-1} N_j} = \frac{\sum_{j=1}^{s-1} j \frac{(k_1\nu)^{j-1}}{(j-1)!}}{\sum_{j=1}^{s-1} \frac{(k_1\nu)^{j-1}}{(j-1)!}} \qquad (19)$$

At the beginning of the reaction 19 gives $\lim_{\nu\to 0} DP_n^\beta = 1$. At large values of ν $\lim_{\nu\to\infty} DP_n^\beta = s - 1$. The number average degree of polymerization of the β-fraction thus varies in the range $1 \leqslant DP_n^\beta \leqslant s - 1$.

The weight average degree of polymerization of the β-fraction is given by

$$DP_w^\beta = \frac{\sum_{j=1}^{s-1} j^2 N_j}{\sum_{j=1}^{s-1} jN_j} = \frac{\sum_{j=1}^{s-1} j^2 \frac{(k_1\nu)^{j-1}}{(j-1)!}}{\sum_{j=1}^{s-1} j \frac{(k_1\nu)^{j-1}}{(j-1)!}} \qquad (20)$$

Thus $\lim\limits_{\nu\to 0} DP_{\mathrm{w}}{}^{\beta} = 1$, and $\lim\limits_{\nu\to\infty} DP_{\mathrm{w}}{}^{\beta} = s - 1$. Hence, $DP_{\mathrm{w}}{}^{\beta}$ varies during the polymerization within the range $1 \leqslant DP_{\mathrm{w}}{}^{\beta} \leqslant s - 1$.

The Fraction and Average Degree of Polymerization of Chains in the α-Form.—The number and weight fractions $P_{\mathrm{n}}{}^{\alpha}$ and $P_{\mathrm{w}}{}^{\alpha}$, respectively, of chains in the α-form (*i.e.*, $j \geqslant s$) may be derived from (16) and (18), remembering that $P_{\mathrm{n}}{}^{\alpha} = 1 - P_{\mathrm{n}}{}^{\beta}$ and $P_{\mathrm{w}}{}^{\alpha} = 1 - P_{\mathrm{w}}{}^{\beta}$.

The number average degree of polymerization of the α-fraction is given by

$$DP_{\mathrm{n}}{}^{\alpha} = \frac{\sum\limits_{j=s}^{\infty} jN_{\mathrm{j}}}{\sum\limits_{j=s}^{\infty} N_{\mathrm{j}}} = \frac{\sum\limits_{j=s}^{\infty} jk_2{}^{j} \int_0^{\nu} \frac{y^{j-s}}{(j-s)!} e^{-\delta y} (\nu - y)^{s-2}\, \mathrm{d}y}{\sum\limits_{j=s}^{\infty} k_2{}^{j} \int_0^{\nu} \frac{y^{j-s}}{(j-s)!} e^{-\delta y} (\nu - y)^{s-2}\, \mathrm{d}y} \tag{21}$$

where the values of N_{j} formulated in (9) have been used. Reversing the order of summation and integration eq. 21 yields

$$DP_{\mathrm{n}}{}^{\alpha} = s + \frac{k_2 \int_0^{\nu} e^{k_1 y}\, y(\nu - y)^{s-2}\, \mathrm{d}y}{\int_0^{\nu} e^{k_1 y} (\nu - y)^{s-2}\, \mathrm{d}y} \tag{22}$$

Introduction of the new variable $x = \nu - y$ leads to

$$DP_{\mathrm{n}}{}^{\alpha} = s + k_2\nu - \frac{k_2 \int_0^{\nu} e^{-k_1 x}\, x^{s-1}\, \mathrm{d}x}{\int_0^{\nu} e^{-k_1 x}\, x^{s-2}\, \mathrm{d}x} \tag{23}$$

At the beginning of the reaction when $\nu \to 0$, $DP_{\mathrm{n}}{}^{\alpha} = s$, since

$$\lim_{\nu\to 0} \frac{\int_0^{\nu} e^{-k_1 x}\, x^{s-1}\, \mathrm{d}x}{\int_0^{\nu} e^{-k_1 x}\, x^{s-2}\, \mathrm{d}x} = \nu$$

At large values of ν

$$\lim_{\nu\to\infty} \frac{\int_0^{\nu} e^{-k_1 x}\, x^{s-1}\, \mathrm{d}x}{\int_0^{\nu} e^{-k_1 x}\, x^{s-2}\, \mathrm{d}x} = \frac{\Gamma(s)/k_1{}^{s-1}}{\Gamma(s-1)/k_1{}^{s-2}} = \frac{s-1}{k_1}$$

Hence

$$\underset{\text{at } \nu\to\infty}{Dp_{\mathrm{n}}{}^{\alpha}} = s + k_2\nu - \frac{k_2}{k_1}(s - 1)$$

The last expression reduces to the well known form

$$DP_{\mathrm{n}}{}^{\alpha} = k\nu + 1, \text{ when } k_2 = k_1$$

The number average degree of polymerization of the α fraction thus varies, during the polymerization, in the range

$$s \leqslant DP_{\mathrm{n}}{}^{\alpha} \leqslant s + k_2\nu - \frac{k_2}{k_1}(s - 1)$$

The weight average degree of polymerization of the α-fraction is given by

$$DP_{\mathrm{w}}{}^{\alpha} = \frac{\sum\limits_{j=s}^{\infty} j^2N_{\mathrm{j}}}{\sum\limits_{j=s}^{\infty} jN_{\mathrm{j}}} = \frac{\sum\limits_{j=s}^{\infty} j^2k_2{}^{j} \int_0^{\nu} \frac{y^{j-s}}{(j-s)!} e^{-\delta y} (\nu - y)^{s-2}\, \mathrm{d}y}{\sum\limits_{j=s}^{\infty} jk_2{}^{j} \int_0^{\nu} \frac{y^{j-s}}{(j-s)!} e^{-\delta y} (\nu - y)^{s-2}\, \mathrm{d}y} \tag{24}$$

where the values of N_{j} for $j = s, s + 1, \ldots, \infty$ were obtained from (9). Equation 24 may be rewritten in the form

$$DP_{\mathrm{w}}{}^{\alpha} = k_2\nu + s + \frac{k_2\nu \int_0^{\nu} x^{s-2}e^{-k_1 x}\, \mathrm{d}x - (k_2{}^2\nu + k_2 s + k_2) \int_0^{\nu} x^{s-1}e^{-k_1 x}\, \mathrm{d}x + k_2{}^2 \int_0^{\nu} x^{s}e^{-k_1 x}\, \mathrm{d}x}{(k_2\nu + s) \int_0^{\nu} x^{s-2}\, e^{-k_1 x}\, \mathrm{d}x - k_2 \int_0^{\nu} x^{s-1}e^{-k_1 x}\, \mathrm{d}x} \tag{25}$$

At the beginning of the reaction, when $\nu \to 0$, the fraction appearing in the right hand side of eq. 25 equals $- k_2\nu$, thus $\lim\limits_{\nu\to 0} DP_{\mathrm{w}}{}^{\alpha} = s$.

At large values of ν

$$\lim_{\nu\to\infty} DP_{\mathrm{w}}{}^{\alpha} = s + k_2\nu + \frac{k_2k_1\nu\Gamma(s-1) - (k_2{}^2\nu + k_2\nu + k_2)\Gamma(s)}{(k_2k_1\nu + k_1 s)\Gamma(s-1) - k_2\Gamma(s)} = s + k_2\nu + 1 - \frac{k_2}{k_1}(s - 1)$$

In the special case in which $k_2 = k_1$ $\lim\limits_{\nu\to\infty} DP_{\mathrm{w}}{}^{\alpha} = k\nu + 2$.

Number and Weight Average Degrees of Polymerization of the Total Polymer Fraction.—The estimation of the number average degree of polymerization of the total polymer fraction may be made by utilizing formulas (16), (19) and (22), since

$$DP_{\mathrm{n}} = P_{\mathrm{n}}{}^{\beta}DP_{\mathrm{n}}{}^{\beta} + P_{\mathrm{n}}{}^{\alpha}DP_{\mathrm{n}}{}^{\alpha} = P_{\mathrm{n}}{}^{\beta}DP_{\mathrm{n}}{}^{\beta} + (1 - P_{\mathrm{n}}{}^{\beta})DP_{\mathrm{n}}{}^{\alpha} \tag{26}$$

Similarly, the weight average degree of polymerization of the total polymers fraction may be derived from the relation

$$DP_{\mathrm{w}} = P_{\mathrm{w}}{}^{\beta}DP_{\mathrm{w}}{}^{\beta} + (1 - P_{\mathrm{w}}{}^{\beta})DP_{\mathrm{w}}{}^{\alpha} \tag{27}$$

using the expression given in (18), (20) and (24).

Evaluation of s, $P_{\mathrm{n}}{}^{\beta}$, $P_{\mathrm{w}}{}^{\beta}$, $P_{\mathrm{n}}{}^{\alpha}$ and $P_{\mathrm{w}}{}^{\alpha}$ from Representative Experimental Data.—Numerical evaluation of the above polymer characteristics is demonstrated below, using the polymerization data for N-carboxy-O-carbobenzoxy-L-tyrosine anhydride and N-carboxy-γ-benzyl-L-glutamate anhydride.

From the kinetic data in Fig. 1 for the polymerization of N-carboxy-O-carbobenzoxy-L-tyrosine anhydride the concentration of monomer M may be derived as a function of the time of polymerization, t, leading to the evaluation of $\nu = \int_0^t M\mathrm{d}t$ at any instant. Thus the curve giving M as a function of t may be transformed into a curve describing M as a function of ν. The values of $\mathrm{d}M/\mathrm{d}\nu$ derived from the latter, when plotted *versus* ν, give a sigmoid curve, the inflection point of which corresponds to

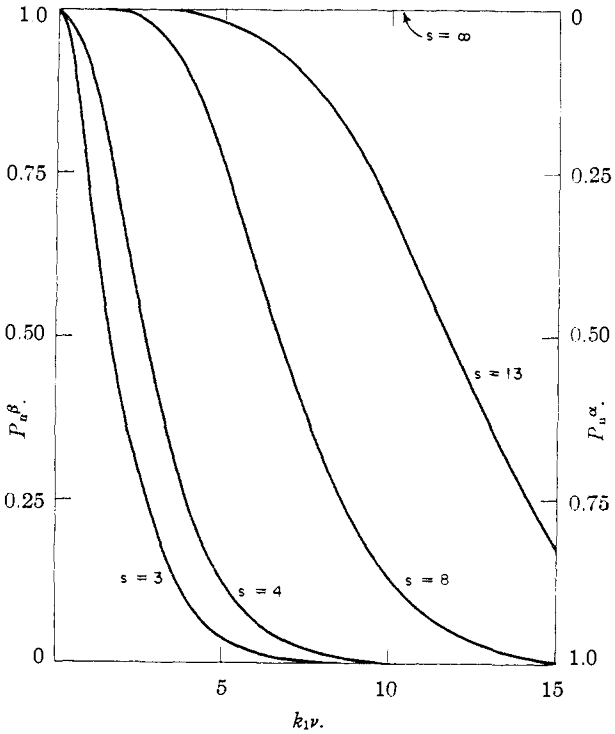

Fig. 2.—Number fraction of peptide chains in the β and α-forms (P_n^β and P_n^α, respectively) as a function of $k_1\nu$ for various values of s.

the transition value of ν at which $d^3M/d\nu^3 = 0$. The value of s may now be evaluated since $s = k_1\nu_{\text{trans.}} + 2$ (see eq. 14a). In the tyrosine experiment $s = 12.8$, since it was found that $\nu_{trans.} = 15.3$ mole liter^{-1} min. and $k_1 = 0.707$ liter mole^{-1} min.$^{-1}$. The fast propagation reaction thus begins when the growing poly-O-carbobenzoxy-L-tyrosine chains reach a degree of polymerization of about 13.

By making use of the data in Fig. 2, it is found that for $s = 13$, $P_n^\beta = 0.58$ when $k_1\nu = 10.8$. Hence 58% of the growing chains are still in the β-form at the transition point, while 42% are in the α-form. The value $P_w^\beta = 0.50$ was derived from the corresponding data of Fig. 3, and the experimental value for $(M_0 - M_t)/I_0$ at the transition point. Thus the polymer fraction at the transition point consists of equal amounts by weight of α- and β-chains. Finally, a calculation of the number and weight average degrees of polymerization of the β-fraction, at the transition point, carried out in accordance with equations 19 and 20, respectively, gave $DP_n^\beta = 9.7$ and $DP_w^\beta = 10.1$.

An analogous calculation to the above also was made for the O-carbobenzoxy-L-tyrosine polymer obtained at the end of the polymerization reaction at which $k_1\nu \simeq 20$. The values obtained for the final polymer were: $P_n^\beta = 0.021$, $P_w^\beta = 0.0059$, $DP_n^\beta = 11.2$ and $DP_w^\beta = 11.2$. Thus the theoretical treatment predicts that approximately 0.6% of the total weight of the polyamino acid synthesized should be in the β form.

A value of $s \simeq 8$ was obtained when the above treatment was applied to a primary amine-initiated polymerization of N-carboxy-γ-benzyl-L-glutamate anhydride in dioxane[3] (where $M_0 = 0.152$ mole liter^{-1}, $M_0/I_0 = 40$, and $k_1 = 0.3$ liter mole^{-1} min.$^{-1}$). At the transition point $k_1\nu_{trans.} = 6.2$, $P_n^\beta = 0.60$, $P_w^\beta = 0.54$, $DP_n^\beta = 5.4$ and $DP_w^\beta = 5.8$. At the end of the polymerization reaction, when $k_1\nu = 10$, the following values were obtained for the β-fraction of the final polymer: $P_n^\beta = 0.125$ and $P_w^\beta = 0.019$. The poly-γ-benzyl-L-glutamate synthesized under the conditions specified[3] should therefore contain approximately 2% by weight of low molecular weight peptide chains with degrees of polymerization up to 8, the remaining 98% consisting of the α-fraction.

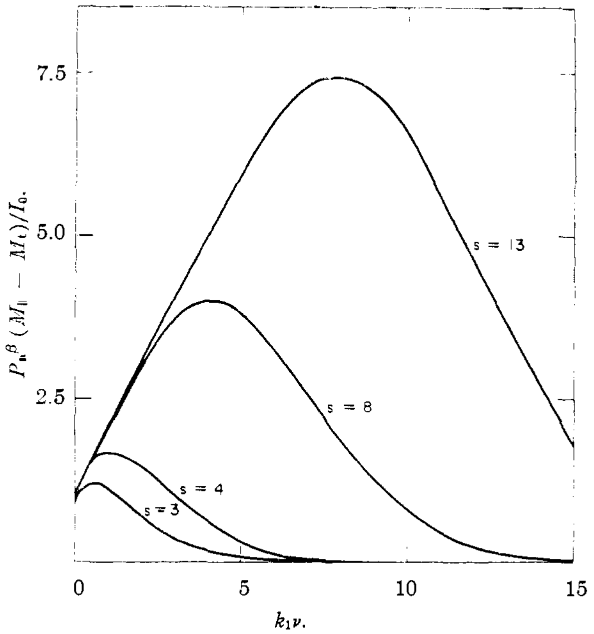

Fig. 3.—Weight fraction P_w^β of chains in the β-form as a function of $k_1\nu$ for various values of s: M_0 and I_0 denote, respectively, the initial concentrations of monomer and initiator, while M_t denotes the concentration of monomer at time t.

Peptide chains in the β and α conformation differ in their infrared absorption spectra[10] as well as in their physicochemical properties. The content of both conformations in a given polyamino acid may therefore be determined using techniques such as infrared analysis or chromatography. In the case of poly-γ-benzyl-L-glutamate it was possible to isolate the β-fraction by extraction with formic acid or alcohol.[11] In view of the theoretical treatment discussed, it would be desirable to estimate the composition of various polyamino acid samples prepared under well specified conditions and to compare the data obtained with those to be expected from the corresponding kinetic experiments.

Appendix I

Evaluation of $\underbrace{\int_0^\nu \ldots \int_0^\nu}_{\substack{j-s+1\\ \text{times}}} \nu^{s-2} e^{\delta\nu} d\nu \ldots d\nu$: The evaluation of this multiple integral can be accomplished by making use of the Laplace transform defined as

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[10] C. H. Bamford, A. Elliott and W. E. Hanby, "Synthetic Polypeptides," Academic Press, Inc., New York, N. Y., 1956, p. 153.

[11] E. R. Blout and A. Asadourian, THIS JOURNAL, **78**, 955 (1956).

$$L\{f(t)\} = \int_0^\infty e^{-pt} f(t)\,\mathrm{d}t = \bar{f}(p) \qquad (28)$$

From (28) one obtains[12] the relation

$$L\left\{\underbrace{\int_0^t\cdots\int_0^t}_{n\text{ times}} f(t)\mathrm{d}t\ldots\mathrm{d}t\right\} = \frac{1}{p^n}\bar{f}(p) \qquad (29)$$

Thus

$$L\left\{\underbrace{\int_0^\nu\cdots\int_0^\nu}_{j-s+1\text{ times}} e^{\delta\nu}\nu^{s-2}\,\mathrm{d}\nu\ldots\mathrm{d}\nu\right\} = \frac{1}{p^{j-s+1}}L\{e^{\delta\nu}\nu^{s-2}\} \qquad (30)$$

However, since

$$L\{t^n\} = \frac{n!}{p^{n+1}} \qquad (31)$$

(see ref. 12), $1/p^{j-s+1}$ may be represented by

$$\frac{1}{p^{j-s+1}} = L\left\{\frac{\nu^{j-s}}{(j-s)!}\right\} \qquad (32)$$

and eq. 30 may be rewritten in the form

$$L\left\{\underbrace{\int_0^\nu\cdots\int_0^\nu}_{j-s+1\text{ times}} e^{\delta\nu}\nu^{s-2}\,\mathrm{d}\nu\ldots\mathrm{d}\nu\right\} = L\left\{\frac{\nu^{j-s}}{(j-s)!}\right\} L\{e^{\delta\nu}\nu^{s-2}\} \qquad (33)$$

Equation 33 may now be transformed into eq. 35 by making use of the convolution lemma (34).[12]

$$\bar{f}(p)\bar{g}(p) = L\left\{\int_0^\nu f(\nu-y)g(y)\mathrm{d}y\right\} \qquad (34)$$

$$L\left\{\underbrace{\int_0^\nu\cdots\int_0^\nu}_{j-s+1\text{ times}} e^{\delta\nu}\nu^{s-2}\,\mathrm{d}\nu\ldots\mathrm{d}\nu\right\} =$$

$$L\left\{\int_0^\nu \frac{y^{j-s}}{(j-s)!}e^{\delta(\nu-y)}(\nu-y)^{s-2}\mathrm{d}y\right\} \qquad (35)$$

By Lerch's theorem[12] we finally obtain

$$\underbrace{\int_0^\nu\cdots\int_0^\nu}_{j-s+1\text{ times}} e^{\delta\nu}\nu^{s-2}\,\mathrm{d}\nu\ldots\mathrm{d}\nu =$$

$$\int_0^\nu \frac{y^{j-s}}{(j-s)!}e^{\delta(\nu-y)}(\nu-y)^{s-2}\mathrm{d}y \qquad (36)$$

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[12] See, for example, F. B. Hildebrand, "Advanced Calculus for Engineers," Prentice–Hall, Inc., New York, N. Y., 1957, p. 52.

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, CARNEGIE INSTITUTE OF TECHNOLOGY, PITTSBURGH, PENNA.]

Complex Formation in the Phosphorus Tribromide–Trimethylamine System and the Phosphorus Trichloride–Trimethylarsine System Phosphorus Nitrogen Chemistry II.[1,2]

BY ROBERT R. HOLMES

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The complexes, $Br_3P\cdot N(CH_3)_3$ and $Cl_3P\cdot As(CH_3)_3$ were prepared and characterized. Dissociation pressures were determined for both solids. The heat of reaction of phosphorus tribromide with trimethylamine was determined from Henry's law constants. The solubility relationship of the solid, $Br_3P\cdot N(CH_3)_3$, as a function of temperature was also established. A value of the heat of solution of the latter in phosphorus tribromide resulted. Nearly ideal liquid behavior was established in the phosphorus trichloride–trimethylarsine system above the "melting" range of $Cl_3P\cdot As(CH_3)_3$. Also the trimethylphosphine–trimethylamine system showed ideal behavior. The data provided an estimation of the bond energy in $Br_3P\cdot N(CH_3)_3$ and allowed a discussion of electronic factors in comparing its stability with the corresponding $Cl_3P\cdot N(CH_3)_3$ complex.

Introduction

The phosphorus–nitrogen linkage postulated to account for the existence of $Cl_3P\cdot N(CH_3)_3$ established previously[1] led to further investigation to ascertain more clearly the nature of the interaction. If the interaction involves the establishment of a phosphorus–nitrogen linkage, variation in the groups attached to either the phosphorus or nitrogen atom should show up in measurements of the relative interaction energies of the resultant molecules. Thus phosphorus tribromide–trimethylamine phase equilibria were studied to estimate the relative importance of decreasing electronegativity on the interaction energy. Going to the opposite situation, trimethylphosphine was considered to ascertain the effect of increasing the electron field around phosphorus. Finally, the effect of using an atom of lesser electron field than nitrogen was examined by studying the trimethylphosphine–trimethylarsine system.

Results

The PBr_3–$(CH_3)_3N$ System.—Additions of $(CH_3)_3N$ to 4.36 mmoles of liquid PBr_3 at 0° resulted in the three phase equilibrium ($C = L + V$) having a constant pressure of 1.6 mm., until a mole ratio of $(CH_3)_3N$ to PBr_3 of 1.00:1.03 was reached; after this the pressure rose sharply. It was noted that nice crystals, several mm. in length could easily be obtained from excess PBr_3.

Vapor pressures of the 1:1 complex were determined from 0 to 28° and are listed in the second row of Table I.

TABLE I

DISSOCIATION PRESSURES OF PHOSPHORUS TRIBROMIDE–TRIMETHYLAMINE

t, °C.	0.0	4.30	8.25	11.90	16.00	19.90	25.1	25.2	28.0
p, mm.	1.6	3.2	5.2	7.6	12.2	19.2	35.1	35.8	48.9
$p(CH_3)_3N$	1.5	3.0	4.8	7.0	11.3	17.7	32.8	33.5	46.0

The procedure followed to obtain the vapor pressures and prove their independence of the relative amount of vapor was similar to that described for the sublimation of $Cl_3P\cdot N(CH_3)_3$.[1]

The third row of Table I lists pressures obtained by subtracting out the contribution due to PBr_3. The latter calculation will be discussed later. However, in this case the correction does not influence

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[1] Presented before the Inorganic Division at the 138th Meeting of the American Chemical Society, New York, N. Y., September, 1960. Previous paper in this series: R. R. Holmes, *J. Phys. Chem.*, **64**, 1295 (1960).