On the Polymerization Mechanism of α-Amino Acid N-Carboxyanhydrides Initiated by Sodium Hydride

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ABSTRACT: We have recently investigated the polymerization of α-amino acid N-carboxyanhydrides using sodium hydride as an initiator. Our results are best explained in terms of the well-established "active monomer" mechanism for these polymerizations.

A question has been raised whether sodium hydride, an even stronger base than sodium methoxide, can initiate polymerization of α-amino acid N-carboxyanhydrides. We recently carried out experiments with purified sodium hydride and the α-amino acid N-carboxyanhydrides (NCA) derived from N^{ϵ}-benzyloxycarbonyl-L-lysine and γ-benzyl-L-glutamic acid. In both cases we found that under aprotic conditions the conversion of these monomers into high molecular weight polypeptides takes place rapidly and quantitatively.

Experimental Section

Polymerization of N^{ϵ}-Benzyloxycarbonyl-L-lysine NCA Initiated by Sodium Hydride. Sodium hydride (6 mg, 50% oil dispersion) was added to a solution of carefully purified N^{ϵ}-benzyloxycarbonyl-L-lysine NCA (0.45 g) in 15 mL of dioxane (monomer-to-initiator molar ratio = 12.5). The solvent was purified and dried according to the literature procedure.[1] Polymerization was monitored by the decrease of the monomer NCA infrared bands at 1790 and 1860 cm^{-1} and by the parallel increase of the amide bands of the polymer at 1650 and 1540 cm^{-1} (Figure 1). The reaction ensued in less than 20 min after the addition of the initiator and the plot of percent conversion vs. time (Figure 2) showed the characteristic autocatalytic behavior,[2] typical of strong base-initiated polymerizations. In dioxane the conversion exceeded 90% in about 150 min. The intrinsic viscosity of the polymer, isolated at the end of the reaction, was 2.1 dL/g in dichloroacetic acid, corresponding to molecular weight greater than 500 000.[3]

Polymerization of γ-Benzyl L-glutamate NCA Initiated by Sodium Hydride. Sodium hydride (0.64 mg, 57% oil dispersion) was added to a solution of highly purified γ-benzyl L-glutamate NCA (0.21 g) (recrystallized twice from CH_2Cl_2/CCl_4, and three times from ethyl acetate/*n*-hexane) in 7 mL of tetrahydrofuran (monomer-to-initiator molar ratio = 50) under an atmosphere of dry nitrogen. Tetrahydrofuran was purified three times by refluxing in the presence of sodium hydride and then distilling under a dry nitrogen atmosphere. All other solvents were dried and purified over calcium hydride. Polymerization was detectable approximately 10 min after addition of the initiator, as monitored by the decrease of the NCA infrared bands at 1790 and 1860 cm^{-1} and by the parallel increase of the amide bands of the polymer at 1650 and 1540 cm^{-1} using a 0.05-mm NaCl liquid cell. In the calculation of percent conversion of the NCA and of percent yield of polymer, the infrared band at 1735 cm^{-1} was used as an internal standard. The kinetic curve of percent conversion of NCA vs. time (Figure 2), which is in good agreement with the kinetic curve of polymer yield vs. time, exhibits the characteristic autocatalytic behavior[2] in tetrahydrofuran as well as in dioxane (see above for N^{ϵ}-benzyloxycarbonyl-L-lysine NCA), reaching greater than 70% conversion in about 150 min. The intrinsic viscosity of the polymer, isolated after 180 min, was 1.55 dL/g in dichloroacetic acid, which corresponds to a molecular weight of 285 000.[4]

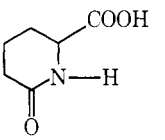
Discussion

Under the polymerization conditions described above, the "active monomer" species is most likely to form and initiate rapid polymerization. Since Seeney and Harwood reported that sodium hydride does not initiate polymerization of NCA's and have questioned the validity of the "active monomer" mechanism, we were led to reexamine many contentions reported in their recent paper.[5]

Seeney and Harwood carried out a series of ^1H NMR and ^{13}C NMR studies of amine–NCA reactions, keeping the ratios of amine to NCA in the range 3:1 to 1:1, and confirmed the well-known observation that reasonably stable carbamate ion adducts are formed in such systems. This reaction has been known for many years; thus in 1950 Bailey[6] isolated adducts of this type and used them in a stepwise peptide synthesis. The reaction was well-documented by Katchalski and Sela[7] and by Bamford and his co-workers.[8,9]

The fact that carbamate-ion adducts are formed when essentially equivalent amounts of amine and NCA are mixed together does not warrant the sweeping conclusion that propagation has only to occur through nucleophilic attack of a carbamate ion intermediate on the NCA as claimed by Seeney and Harwood.[5] Such carbamate intermediates have, in fact, been prepared by Kopple,[10] who had earlier postulated[11] that polymerization occurred via such an intermediate. However, Kopple found the rate of decarboxylation of the mixed carbamic–carboxylic anhydride to be too slow to account for the rate of polymerization observed using strong base (tertiary amine) initiation. He therefore concluded that these mixed anhydride intermediates do not participate in the rapid polymerization of NCA's.

An alternative mechanism is needed to account for these reactions. Lack of incorporation of the initiator during the polymerization of γ-benzyl glutamate NCA initiated by ^{14}C-labeled sodium methoxide[12,13] and ^{14}C-labeled sodium benzylcarbamate[13,14] or 9-fluorenylpotassium in dioxane[12] under rigorous aprotic conditions provides strong support for the "active monomer" mechanism. Using strong base initiators with δ-benzyl-α-aminoadipic acid NCA under stringently dry, aprotic conditions, Choi and Goodman[15] showed that compound I can be isolated as a major product:



I

It is most reasonable to conclude that compound I is formed by the abstraction of a proton from the amide N–H bond of the NCA, which then intramolecularly attacks the ester group. Formation of this product is also consistent with the "active monomer" mechanism of polymerization.

The "active monomer" mechanism of polymerization must lead to the formation of a polymer with an N-carboxyanhydride terminal group, structure II. The presence of such N-

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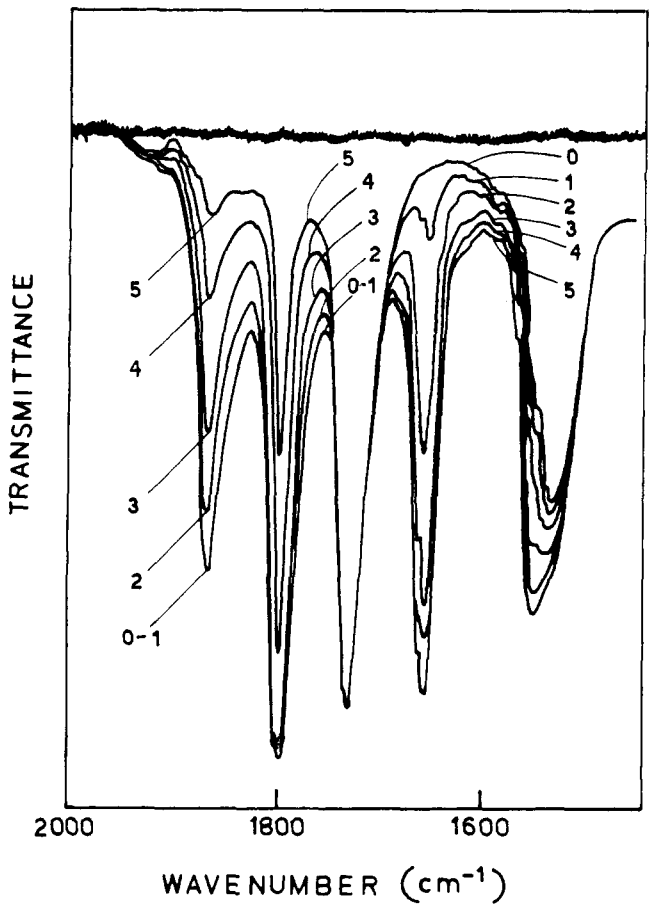
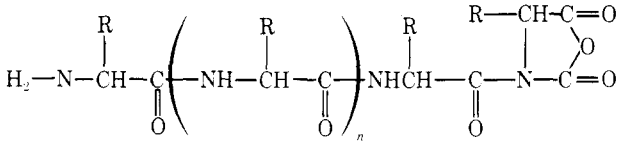


Figure 1. Polymerization of N^{ϵ}-benzyloxycarbonyl-L-lysine NCA in dioxane initiated by sodium hydride. Infrared spectra recorded at various reaction times: initial (spectrum 0); 20 min (spectrum 1); 45 min (spectrum 2); 70 min (spectrum 3); 100 min (spectrum 4); 130 min (curve 5). The monomer to initiator molar ratio was 12.5.



II

carboxyanhydride terminal groups in the polymers prepared by aprotic basic initiation can be deduced from the post-polymerization properties of the polymers.[15,17] Addition of fresh NCA does not lead to an increase in the molecular weight of the pre-existing polymer. From this observation, we deduce that the polymer is not the attacking species. Addition of a radioactive primary amine after completion of the polymerization leads to incorporation of radioactivity in the polymer. The amount of incorporated radioactivity is substantially reduced when the polymer is stored or precipitated and redissolved before addition of the radioactive primary amine. These results are easily explained by the presence of an active NCA terminal group on the polymer.

Experiments involving ^{14}C-labeled sodium methoxide were repeated by Seeney and Harwood.[5] Not surprisingly, they found radioactivity in the isolated polypeptide since they employed radioactive methoxide in radioactive methanol. The latter can react with NCA monomer and yields a low molecular weight polymer with a radioactive terminal group.[7,18] Alternatively, the radioactive methanol can open the ring at the terminus of structure II (above) and thus yield high molecular weight polymer containing radioactivity.

Seeney and Harwood also remark that tertiary amines fail to initiate polymerization of NCA's in the absence of "impurities" and in support cite the work of Ledger and Stewart[19] and Kricheldorf.[20] The former authors were mainly concerned with the preparation of polypeptides from NCA's and their observations can hardly be interpreted to imply lack of initiation by tertiary bases, while Kricheldorf's work dealt with the NCA's of β-amino acids. Seeney and Harwood do not specify the type of NCA or the conditions used in their experiments. It is a well-established experimental fact that N-unsubstituted NCA's and N-substituted NCA's in the presence of tertiary bases show types of behavior qualitatively different. Under conditions which lead to rapid polymerization of the former, the latter (e.g., sarcosine or L-proline NCA's) are unaffected for long periods.[21] This matter was studied and debated exhaustively over 20 years ago, to an extent which should have eliminated the need for consideration of adventitious impurities. Another fact, not considered by Seeney and Harwood, is that solvent properties have a profound effect on the rates of polymerization initiated by tertiary bases and also on the nature of the products; examples have been given by Bamford et al.[22] Low rates of reaction in a solvent of low dielectric constant such as dioxane in no way imply that "impurities" are necessary for initiation.

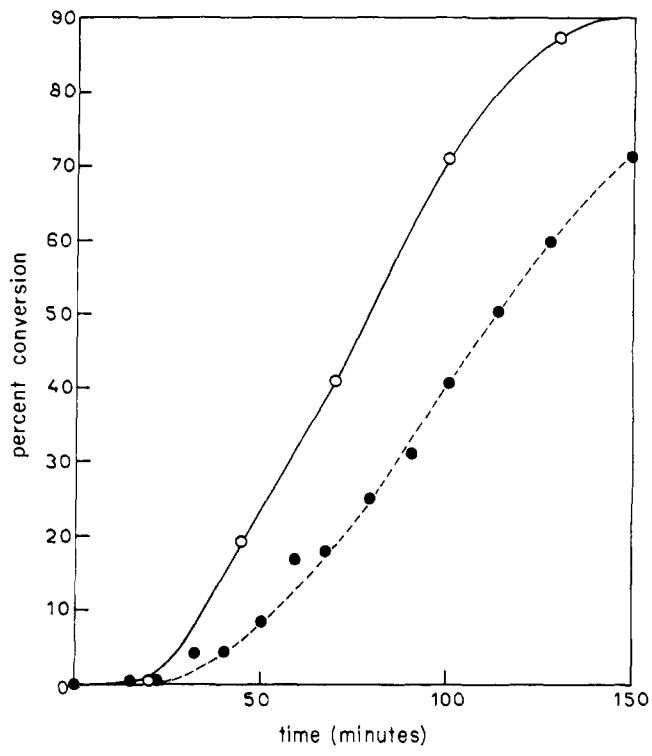


Figure 2. Kinetic curves of conversion (%) vs. time for the sodium hydride initiated polymerization of N^{ϵ}-benzyloxycarbonyl-L-lysine NCA in dioxane (—○—○—) and of γ-benzyl L-glutamate NCA in tetrahydrofuran (—●—●—). (The curve for N^{ϵ}-benzyloxycarbonyl-L-lysine NCA was calculated from the spectra of Figure 1. The curve for γ-benzyl L-glutamate NCA was calculated from infrared spectra recorded at the following reaction times: initial; 15 min; 22 min; 32 min; 40 min; 50 min; 59 min; 67 min; 79 min, 90 min; 100 min; 113 min; 127 min; 149 min.)

Conclusions

It is well established that, under rigorous aprotic conditions of monomer and solvent, polymerization of NCA is initiated by sodium hydride, most likely via the "active monomer" mechanism originally proposed by Bamford and Block,[23] and subsequently modified by Szwarc.[24] The observations reported by Seeney and Harwood do not invalidate the "active monomer" mechanism; in fact, some of their results may be used in its support.

Much work remains to be carried out in the field of NCA polymerization. The physical factors such as the conformation of the growing chain,[24] effects of heterogeneity and monomer absorption on the active ends of growing chains,[25] effects of counterions, and the modes of molecular weight distributions of polymers all require further elucidation.[26]

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(26) **Note Added in Proof:** After submission and acceptance of our manuscript, a paper appeared by H. R. Kricheldorf, *Makromol. Chem.*, **178**, 1959–1970 (1977), in which he clearly demonstrated that the "active monomer" mechanism is valid for NCA polymerizations. In earlier papers, he showed the formation of NCA anions by the essentially quantitative *N*-silylation and *N*-sulfenylation of NCA's [H. R. Kricheldorf, *Angew. Chem.*, **85**, 86 (1973) and H. R. Kricheldorf, and M. Fehrle, *Chem. Ber.*, **107**, 3533–3547 (1974)].

Cationic Polymerization of 2-Alkoxy-2-oxo-1,3,2-dioxaphosphorinanes (1,3-Propylene Alkyl Phosphates)

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ABSTRACT: Kinetics, thermodynamics, and mechanism of polymerization of 1,3-propylene alkyl phosphates, the six-membered cyclic esters of phosphoric acid, have been investigated. The reaction was performed in bulk, using cationic initiators such as salts of Ph_3C^+ with stable anions (PF_6^-, AsF_6^-). The kinetics of polymerization was resolved and the elementary steps were described. Propagation is reversible, as in case of the previously studied 1,3-propylene methyl phosphate where the enthalpies and entropies of the propagation–depropagation equilibria gave a linear isoequilibrium plot. ΔH_p and ΔS_p both increase with the size of the exocyclic groups. Only the polymerization of 1,3-propylene methyl phosphate is exothermic; polymerizations of monomers with larger exocyclic groups are endothermic and possible because of the positive entropy change. Apparently, the mobility of the large exocyclic groups, having restricted rotation in monomers, increases in polymers and the gain in the rotational entropy, also not too large (a few entropy units), is sufficient to shift the propagation–depropagation equilibrium to the polymer side. ^1H and ^{31}P NMR spectra indicate that polymers are linear with cyclic end groups, formed because of the extensive chain transfer to monomer of the positively charged exocyclic groups from the growing centers.

In our previous paper, describing the kinetics and thermodynamics of the cationic polymerization of 2-methoxy-2-oxo-1,3,2-dioxaphosphorinane (1,3-propylene methyl phosphate) (1), we stressed the importance of the chain transfer to monomer (eq 1) responsible for the formation of polyesters of limited polymerization degrees.[1] The competition between chain propagation and chain transfer is shown in the Scheme I below.

Thus, for both propagation and transfer, involving nucleophilic attack of the phosphoryl oxygen atom, there is a common transition state proposed, and the mechanism of transfer, illustrated above, leads to the formation of macromolecules with cyclic end groups. The presence of these end groups was confirmed by ^1H, ^{31}P, and ^{13}C NMR.[1,2]

For the methoxy exocyclic group the polymerization degrees of polymers prepared at 100 °C were close to 10, indicating that the ratio k_p/k_{tr} (eq 1) is of the same magnitude.

According to the proposed structure of the transition state (eq 1), it would be necessary to destabilize the carbenium ion of the exocyclic group, partially developed in the transition state, in order to increase the k_p/k_{tr} ratio and in this way to increase the polymerization degree.

We report in this paper on the polymerization of 2-alkoxy-2-oxo-1,3,2-dioxaphosphorinanes of the general formula: