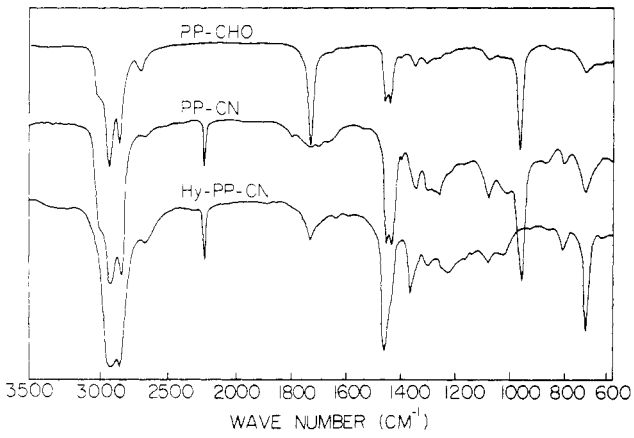

Figure 1. Infrared spectra for PP–CHO (20), PP–CN (20), and Hy–PP–CN (20).

formyl groups present at the completion of the hydrogenation were reconverted to the nitrile as follows.

$$\text{PP}-\text{CN} \xrightarrow{H_2} \text{Hy}-\text{PP}-\text{CN} + \text{Hy}-\text{PP}-\text{CH}=\text{NH}$$
$$\text{Hy}-\text{PP}-\text{CH}=\text{NH} \xrightarrow[+H_2O]{-NH_3} \text{Hy}-\text{PP}-\text{CHO} \quad (3)$$

The hydrogenated polymer (8 g) was dissolved in a mixture of *p*-xylene (500 ml) and pyridine (100 ml) at 80° under nitrogen, and hydroxylamine hydrochloride (1 g) was added. Acetic anhydride (100 g) was added to this hot mixture in portions. After the resulting mixture was heated at 110° for 1 hr with stirring, the product was isolated, purified, and dried in the usual manner. The polymer was purified by reprecipitation from *p*-xylene–pyridine solution into methanol and then dried at 40° *in vacuo* for 15 hr.

Polymer characterizations were carried out on the basis of elemental analysis, Gel Permeation Chromatography (gpc), and infrared spectroscopy (ir). The results are shown in Tables II and III and Figure 1, respectively. Table II shows good agreement between calculated and observed values of carbon, hydrogen, and nitrogen for all the derivatives.

Table III indicates a slight broadening in molecular weight distribution in going from the original PP to the most highly substituted derivative PP–CN (20). The effect is very small, however, and indicates that no significant backbone degradation has occurred as a result of the reaction sequences described above. Gpc measurements were also carried out on the Hy–PP–CN derivatives at 135° using dichlorobenzene as solvent. The resolution was too poor under these conditions to permit meaningful comparisons to be made. This poor resolution may stem from association of the polymer in solution or interaction of the polymer with the column due to the presence of the polar nitrile groups.

Figure 1 presents a comparison of the ir spectra of the PP–CHO, PP–CN, and Hy–PP–CN derivatives. There is some evidence for the presence of a small amount of acylated oxime groups (CH=N–$OCOCH_3$) in PP–CN and Hy–PP–CN (band at around 1740 cm^{-1}). This is presumably an intermediate in the dehydration of the oxime with acetic anhydride. In addition, the carbonyl stretching band at 1740 cm^{-1} is more prominent in Hy–PP–CN than in PP–CN. This could be due to the acylation of small amounts of hydroxyl side groups formed during hydrogenation by acetic anhydride. A suggested sequence is presented in eq 4.

$$\text{PP}-\text{CN} \xrightarrow{H_2} \text{Hy}-\text{PP}-\text{CN} + \text{Hy}-\text{PP}-\text{CH}=\text{NH} \xrightarrow[+H_2O]{-NH_3} \text{Hy}-\text{PP}-\text{CHO} \quad (4)$$
$$\text{Hy}-\text{PP}-\text{CHO} \xrightarrow{H_2} \text{Hy}-\text{PP}-\text{CH}_2\text{OH} \xrightarrow{Ac_2O} \text{Hy}-\text{PP}-\text{CH}_2\text{OCOCH}_3$$

It may be concluded that the methods outlined in this note lead to CN derivatives of the starting PP which are linear and of approximately the same mojecular weight and distribution as the PP itself. The PP–CN derivatives contain small quantities of acylated oxime side groups and acetate side groups as a result of side reactions. In addition, similar experiments with anionically polymerized polybutadiene lead to essentially the same results as the PP, no gelation occurs in contrast to the earlier study cited in the introduction. Also, infrared measurements indicate the absence of hydroxyl groups in the PP–CHO prepared by the present technique. The hydroformylation reaction with the cobalt catalyst leads to appreciable quantities of hydroxyl groups in the product.

Table III
Gpc Data

Sample	Solvent[a]	$\bar{A}_n$, Å	$\bar{A}_w$, Å	$\bar{A}_w/\bar{A}_n$
PP	THF	4700	9,000	1.9
PP–CN (10)	THF	4300	8,400	2.0
PP–CN (15)[b]	THF	5000	8,700	1.7
PP–CN (20)	THF	3300	8,200	2.5
Hy–PP	TCB	6100	10,800	2.8

[a] For THF as solvent: room temp; column pore sizes of 10^6, 10^5, 10^4, 10^3 Å. For TCB as solvent: 135°; column pore sizes of 10^6, 10^5, 10^4, 10^3 Å. [b] PP–CN (15) means PP having about 12.5 mol % of pendant nitrile group.

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The Far-Infrared Spectrum of Collagen

While the near and middle infrared absorption spectra of collagen have been assiduously studied (see ref 1 for a review), the far infrared region, *i.e.*, the region below *ca.* 400 cm^{-1}, has not received much attention. We report here the spectra of native, partially denatured and fully denatured collagen in the range 400–240 cm^{-1} and comment on their use toward the determination of the relative amount of denatured collagen in the solid state.

Collagen was extracted from rat tail tendon with 0.05 *M* acetic acid, using a method similar to that of Piez, Lewis, Martin, and Gross.[2] A specimen of collagen extracted from bovine hide by H. I. Sinnamon and coworkers (United States Department of Agriculture, Philadelphia) and purified in our laboratory was also used as a control. (Spectra obtained with the two collagen specimens were identical.) A solution of denatured collagen was prepared from the

Table I
Relative Amount of Native Collagen in Various Specimens

Specimen description	Far-infrared data A_{345}/A_{1450} (± 3%)	% H	Mid-infrared data A_{1235}/A_{1450}	% H
Native collagen, cast at 23°	0.295	100	1.38	100
Gelatin, cast at 4°	0.227	76 ± 12	1.15	71 ± 9
Gelatin, cast at 23°	0.158	46 ± 10	0.94	44 ± 10
Gelatin, cast at 60°	0.054	0	0.59	0

above by heating at 60° over 15 min. Films of native collagen, ranging in thickness from 5 to 50 μ, were cast from 0.05 M acetic acid solution at 23° while films of collagen varying in extent of denaturation were cast from the solution of denatured collagen at 4, 23, and 60°. The native character of films cast from 0.05 M acetic acid native collagen solution at 23° has been confirmed by detailed analysis of the tensorial components of the optical activity of such films.[3] Spectra were obtained with Perkin-Elmer infrared spectrophotometers Models 521 and 621. A filter change occurs with the 621 instrument near 295 cm^{-1} and for this reason the observed absorbance in the range 300–200 cm^{-1} requires a correction in the form of a vertical shift. The magnitude of the required correction as well as confirmation of the absorbance values observed with the 621 instrument were both conveniently obtained by repeating the measurements with the 521 instrument. Results obtained with the two instruments agreed within 5% (except in the range 250–200 cm^{-1} which is not available with the 521 instrument; also, data obtained in this region with the 621 instrument were poorly reproducible).

The results show that native collagen has an absorption peak at 345 cm^{-1} (see spectrum A in Figure 1). In addition, our data in the region 300–250 cm^{-1} suggest the possibility that several weak absorption bands appear in this region but the resolution of our instruments does not justify a more definitive statement about the location or nature of these bands.

There is, however, no doubt that the 345-cm^{-1} band of collagen originates in the triple-helical (tertiary) structure of the molecule. This conclusion emerges unavoidably from the observation (spectrum D in Figure 1) that the band is effectively abolished when the specimen is prepared by casting at 60°; the latter procedure is the method, well-known from the X-ray diffraction work of Bradbury and Martin,[4] of preparing "hot-cast" gelatin, a protein which has the amino acid sequence (primary structure) of native collagen but which lacks any higher level of structural order. Bradbury and Martin[4] also observed that films cast at 23° from a gelatin solution ("cold-cast" gelatin) gave an X-ray diffraction pattern which was similar, though not identical, to that of native collagen. Spectrum C in Figure 1, obtained with a film cast at 23° from a solution of denatured collagen, indeed shows a peak absorbance which is intermediate between that of the native film and that of the fully denatured film. In the course of our work we discovered that it is possible to prepare a form of gelatin which is less denatured than the "cold-cast" gelatin of Bradbury and Martin[4] by casting a gelatin solution at 4°. The spectrum obtained thereby is presented in Figure 1 (spectrum B).

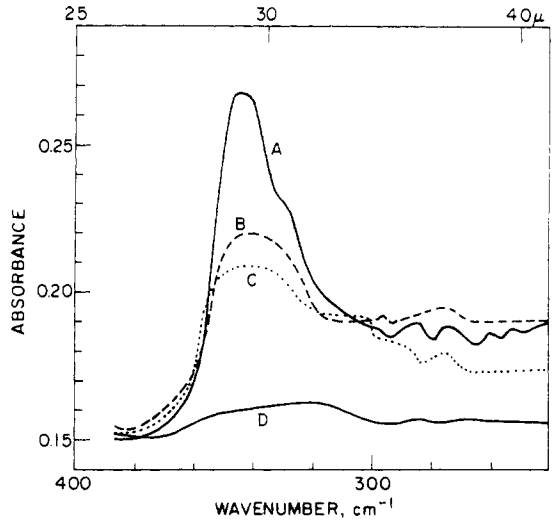

Figure 1. The spectra of native, partly denatured, and fully denatured collagen films in the region 400–240 cm^{-1}: A, native collagen, cast at 23°; B, denatured collagen (gelatin) cast at 4°; C, denatured collagen, cast at 23° ("cold-cast" gelatin); D, denatured collagen, cast at 60° ("hot-cast" gelatin). All films cast from 0.05 M acetic acid solution.

Values of the peak absorbance at 345 cm^{-1}, A_{345}, were obtained by use of a straight base-line tangent to the troughs on either side of the band. Differences in specimen thickness were corrected for by dividing values of A_{345} by the peak absorbance at 1450 cm^{-1}; the latter band serves as a useful internal standard since it has been shown[5] to be invariant with gross changes in the relative extent of denaturation. The ratio $R = A_{345}/A_{1450}$ was accordingly considered to be a measure of the apparent helix content (per cent of maximum possible helicity or % H) by arbitrarily taking R eqivalent to 0% H for hot-cast gelatin (cast at 60°C) and to 100% H for native collagen and by using linear interpolation to assay for specimens possessing R values intermediate between these two extremes. The results appear in Table I.

In order to test the validity of linear interpolation we compared the values of % H obtained above with values obtained by use of a different band in the collagen spectrum, the amide III band occurring at 1235 cm^{-1}, which has been shown[1] to be very sensitive to the presence of the tertiary structure of native collagen. The results obtained by use of the two bands, one in the mid infrared and the other in the far infrared, show good agreement (Table I). Other results[5] show agreement of the % H values obtained by infrared spectroscopy with values obtained by use of optical rotatory dispersion.

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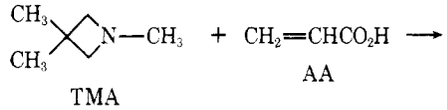
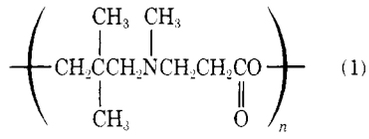
Polymerization *via* Betaine. V.[1] Alternating Copolymerization of 1,3,3-Trimethylazetidine with Acrylic Acid. A Novel Method for the Preparation of Amine-Ester Type Polymer

This communication describes an alternating copolymerization of 1,3,3-trimethylazetidine (TMA) with acrylic acid (AA) to give an amine-ester type polymer **1**. Recently, we



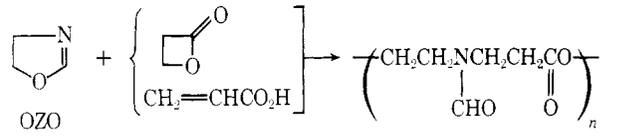
TMA AA



(1)

1

have reported a new type of alternating copolymerizations of cyclic imino ethers, *e.g.*, 2-oxazoline (OZO) and 2-benzyliminotetrahydrofuran (BIT), with β-propiolactones (β-PL) and with AA to produce alternating copolymers of amide-ester type structures, **2** and **3**, respectively (eq 2 and 3).[1-4] A mechanism of the "polymerization *via* betaine" has



OZO

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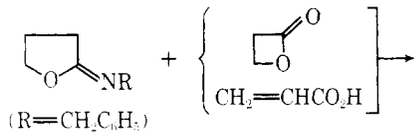
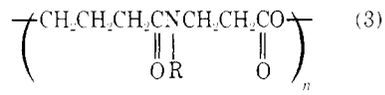
(2)



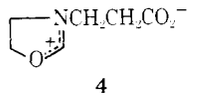
BIT



(3)

3

been proposed for these copolymerizations, *i.e.*, **4** and **5** are considered to be the key intermediates in the alternating copolymerizations of OZO-βPL and OZO-AA (eq 2)[2-4] and BIT-βPL and BIT-AA (eq 3),[1] respectively. In both reactions (eq 2 and 3) cyclic imino ethers of OZO and BIT serve to provide a cationic site of betaines of **4** and **5**.



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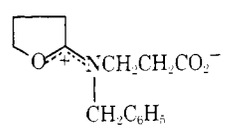

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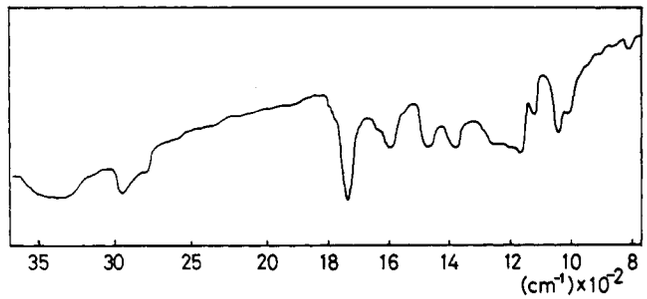

Figure 1. Ir spectrum of the TMA-AA copolymer (KBr).

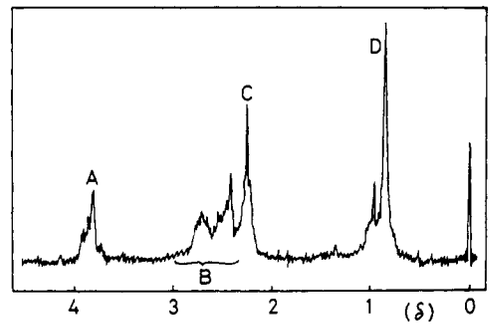

Figure 2. Nmr spectrum of the TMA-AA copolymer ($CDCl_3$).

As an extension of our studies on the copolymerization *via* betaine, we employed TMA as a comonomer providing a cationic site of the betaine components. Now, we disclose a novel type of alternating copolymerization of TMA with AA (eq 1). Cationic homopolymerization and its kinetics of TMA have recently been carried out by Schacht and Goethals.[5,6] However, the copolymerization of TMA has not been reported so far.

An equimolar mixture (3.3 mmol each) of TMA and AA in 1.0 ml of acetonitrile was placed in a sealed tube under nitrogen and kept at 80° for 3.5 hr. Copolymerization took place without added initiator. The reaction mixture was then poured into a large excess of diethyl ether to precipitate the polymeric material. The gummy polymer was separated by filtration, dissolved in chloroform, reprecipitated by pouring the solution into an excess amount of diethyl ether, and dried *in vacuo*. The yield was 0.51 g (89%). The structure of the copolymer was determined by nmr, ir, elemental analysis, and an alkaline hydrolysis experiment.

The ir spectrum of the copolymer (Figure 1) shows absorptions at 1735, 1170, and 1040 cm^{-1}, which indicate the presence of an ester group. Figure 2 shows the nmr spectrum of the copolymer. A signal at δ 3.8 (peak A) is ascribed to methylene protons of $-OCH_2-$. Signals at δ 3.0–2.2 (B) are due to three kinds of methylene protons of $-CH_2NCH_2CH_2CO_2-$. Signals at δ 2.2 (C) and 0.9 (D) are assigned respectively to methyl protons of NCH_3 and $C(CH_3)_2$. The relative intensity of signals A:(B + C):D was 2.0:9.0:6.0. Furthermore, signals due to neither olefinic protons nor carboxylic acid proton were observed. These spectral data strongly support that the structure of the TMA-AA copolymer is **1**. The results of the elemental analysis show the 1:1 composition of TMA and AA.

Anal. Calcd for $(C_9H_{17}NO_2)_n$: C, 63.13; H, 10.00; N, 8.18. Found: C, 62.85; H, 9.95; N, 8.30.

The alkaline hydrolysis experiment further confirmed the copolymer structure **1**. The copolymer (50 mg) was dissolved in 1.0 ml of a 10% solution of NaOH in D_2O and kept at 65° for 1 hr. The reaction mixture was then subjected to nmr measurement. The sole product was the sodium salt of