Journal of Macromolecular Science, Part A
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597274

A Study of the Molecular Weight Distribution of Poly(Amino Acid)s Synthesized by Diphenyl Phosphoryl Azide

Jian-Zhong Yang[a]; Min Wang[a]; Raphael M. Ottenbrite[a]
[a] Department of Chemistry, Virginia Commonwealth University Richmond, Virginia

To cite this Article Yang, Jian-Zhong , Wang, Min and Ottenbrite, Raphael M.(1993) 'A Study of the Molecular Weight Distribution of Poly(Amino Acid)s Synthesized by Diphenyl Phosphoryl Azide', Journal of Macromolecular Science, Part A, 30: 8, 503 — 516
To link to this Article: DOI: 10.1080/10601329308009426
URL: http://dx.doi.org/10.1080/10601329308009426

J.M.S.—PURE APPL. CHEM., A30(8), pp. 503–516 (1993)

A STUDY OF THE MOLECULAR WEIGHT DISTRIBUTION OF POLY(AMINO ACID)S SYNTHESIZED BY DIPHENYL PHOSPHORYL AZIDE

JIAN-ZHONG YANG, MIN WANG, and RAPHAEL M. OTTENBRITE*

Department of Chemistry
Virginia Commonwealth University
Richmond, Virginia 23284

ABSTRACT

Direct polycondensations of β-benzyl-L-aspartate (Asp.Bz) and γ-benzyl-L-glutamate (Glu.Bz) were carried out in the presence of diphenyl phosphoryl azide (DPPA) as a condensation agent and triethyl amine (TEA). Poly(amino acid)s were obtained by this convenient approach whose structure was confirmed by IR and ^{1}H-NMR spectroscopy. The effects of the monomer concentration, the polymerization time and temperature, the ratios [DPPA]/[monomer] and [TEA]/[monomer], and the solvent used on the molecular weight distribution of the polymer were studied. When the monomer concentrations were higher than 0.2 g/mL, poly(Asp.Bz) with a bimodal molecular weight distribution was obtained (M_w of 37,000 and M_w/M_n of 1.68). The polycondensations carried out in THF or in bulk provided the highest molecular weight ($M_w \simeq$ 40,000). Several other amino acids were also polymerized by DPPA.

INTRODUCTION

Synthetic poly(amino acid)s, the analogues of protein and natural peptides, are expected to be useful as biomaterials since the monomers are naturally occurring and the polymers are usually nontoxic substances [1, 2]. Currently, however, only a few practical applications have been developed for these polymers, partly because of the preparation difficulties.

The most popular approach for the synthesis of poly(amino acid)s is the NCA (α-amino acid *N*-carboxyanhydride) method which was first discovered in 1906 by Leuchs et al. [3–5]. This method has been used successfully for the polymerizations of several α-amino acids to obtain good yields of high molecular weight polymer [6–8]. However, this is not a facile method since NCAs are usually very unstable and the polymerization of NCAs requires strict anhydrous conditions. The synthetic polycondensations of amino acids were also carried out thermally [9–24]. It was noted, however, that thermally prepared polymers have problems with structure control and molecular weight. These factors influence the reproducibility as well as the properties of the products.

The first specific polypeptide synthesis involved the Merrifield active ester method. This procedure requires repeated protection, activation, coupling reaction, and deprotection reactions [25, 26]. Therefore, it is too complicated to be used for very high molecular weight polymer synthesis. A sophisticated biosynthesis of protein materials has recently been developed by Fournier et al. [27]. Novel classes of protein-based materials have been created by this genetic engineering, and multigram quantities have been reported.

Direct polycondensation of dicarboxylic acids and diols or diamines with phosphorus compounds as condensation agents has been used successfully to prepare polyesters and polyamides [28–40]. Recently, the synthesis of racemization-free poly(amino acids) by direct polycondensation of α-amino acids using diphenyl phosphoryl azide (DPPA) [41–43] as the condensation agent was proposed by Nishi et al. [44–48]. Several poly(amino acid)s have been prepared by stirring a solution of an amino acid in the presence of DPPA and a tertiary amine.

A major problem associated with the DPPA method is that relatively low molecular weight products are obtained. Furthermore, no detailed information has been reported with respect to the molecular weight distribution. We are interested in investigating this new synthetic method in more detail with respect to it being more convenient than the other procedures for poly(amino acids) synthesis in large quantities.

In the present study, direct polycondensations of β-benzyl-L-aspartate (Asp.Bz) and γ-benzyl-L-glutamate (Glu.Bz) in the presence of DPPA and triethyl amine (TEA) as a base were carried out to ascertain optimal conditions for the preparation of high molecular weight polymers. The effects of various parameters on the molecular weight distribution of the polymer obtained were investigated. The molecular weight distribution of the polymers were evaluated by gel-permeation chromatography (GPC). The polymers were characterized by IR and NMR spectroscopy and compared with those prepared by the NCA method. Several other amino acids, including L-phenylalanine, L-tyrosine, L-valine, and L-isoleucine, were also polymerized by DPPA for comparison.

EXPERIMENTAL

Materials

All major chemicals were purchased from Aldrich Chemical, Inc. β-Benzyl-L-aspartate (Asp.Bz) was prepared by esterification of L-aspartic acid with benzyl alcohol in the presence of concentrated hydrochloric acid (36.5%) at 60°C. The

ester product was purified by recrystallization twice from hot water, mp 215–217°C. Similarly, γ-benzyl-L-glutamate (Glu.Bz) was prepared from L-glutamic acid and benzyl alcohol in the presence of concentrated hydrobromic acid (48%), mp 172–173°C. Diphenyl phosphoryl azide (DPPA) was used without further purification. Triethylamine (TEA) was distillated before use. Solvents for polymerization were purified by common procedures.

Polymerization

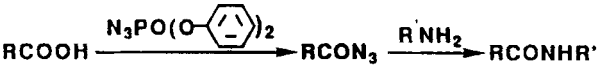
Polycondensations of amino acids were carried out according to the following procedure: 0.25 g (2.24 mmol) of Asp.Bz or 0.26 g (2.24 mmol) of Glu.Bz was mixed with 1 mL of *N,N'*-dimethylformamide (DMF). The system was stirred while it was being cooled in an ice-water bath. To the mixture, 0.63 mL (2.91 mmol) of DPPA and 0.72 mL (5.15 mmol) of TEA were added at 0°C. The reaction mixture was continuously stirred while it was kept in an ice bath for 1 hour, followed by room temperature for 2 days. The polymerization mixture was originally a heterogeneous system; after 2 hours at room temperature the reaction solution became clear. After 2 days the polymer was precipitated in 300 mL water, collected by filtration, washed with water, and then dried in vacuo. The polymers obtained were fractionated by precipitation techniques from a THF/methanol system and by GPC column using Bio-Beads.

Characterization

^{1}H-NMR spectra were recorded on a GE NMR QE-300 (300 MHz) in DMSO-d_6 at room temperature. FT-IR measurements were performed on a Perkin-Elmer 1600 FT-IR spectrometer. The IR spectra of the monomers were measured in KBr disks. The IR spectra of the polymers were measured between NaCl plates. The molecular weights of the polymers were determined on a Perkin-Elmer LC-100 GPC-HPLC [column: μStyragel 10^3 Å, Waters, Inc., 7.8 × 300 mm; mobile phase: tetrahydrofuran (THF); detection: UV detector at 250 nm] at 30°C. Calibration was carried out by using polystyrene standards.

RESULTS AND DISCUSSION

Polycondensation of amino acids using diphenyl phosphoryl azide (DPPA) has been considered to follow a racemization-free procedure as reported by Shioiri et al. for peptide preparation [41–43]. Initially the azide group of DPPA attacks the carboxylic position of the amino acid to give a carboxylic acid azide intermediate. The amino component of the amino acid then reacts with this intermediate to form a peptide amide bond.



It is understandable that for each mole of reacting —COOH on the amino acid, at least 1 mol DPPA is required as the condensation agent. On the other

TABLE 1. Effect of Monomer Concentration on the Polymerization of Asp.Bz by DPPA in DMF at Room Temperature: [DPPA]/[M] = 1.3; [TEA]/[M] = 2.3

		GPC data			
[M], g/mL	Yield, %	M_w,[a] Fraction 1	M_w,[a] Fraction 2	M_w/M_n, Fraction 1	M_w/M_n, Fraction 2
0.025	71.5	–	5800 (100)	–	4.15
0.033	74.7	–	3500 (100)	–	3.50
0.05	67.2	–	5600 (100)	–	5.11
0.10	63.2	–	3400 (100)	–	3.70
0.20	85.4	30,000 (60.7)	1130 (39.3)	1.84	1.13
0.50	86.5	22,200 (59.4)	1000 (40.6)	2.22	1.08
1.0	97.6	27,300 (71.4)	920 (28.6)	1.81	1.05

[a]The value in parentheses is the molar percentage calculated from the GPC curve.

hand, at least 2 mol triethylamine (TEA) is necessary to neutralize the by-products, i.e., hydrazoic acid (HN_3) and diphenyl phosphoric acid.

Effect of Monomer Concentration

The direct polymerizations of Asp.Bz and Glu.Bz were carried out in the presence of DPPA ([DPPA]/[monomer] = 1.3) and TEA ([TEA]/[monomer] = 2.3). The results for the polymerization carried out in DMF at room temperature for 2 days are listed in Tables 1 and 2. Poly(Asp.Bz) and poly(Glu.Bz) were obtained in yields of 98 and 88%, respectively.

TABLE 2. Effect of Monomer Concentration on the Polymerization of Glu.Bz by DPPA in DMF at Room Temperature: [DPPA]/[M] = 1.3; [TEA]/[M] = 2.3

		GPC data			
[M], g/mL	Yield, %	M_w,[a] Fraction 1	M_w,[a] Fraction 2	M_w/M_n, Fraction 1	M_w/M_n, Fraction 2
0.05	85.7	–	3870 (100)	–	2.81
0.10	84.9	13,800 (74.1)	2540 (25.9)	2.78	2.00
0.20	82.2	16,200 (69.4)	1420 (30.6)	1.68	1.05
0.50	85.7	17,300 (71.8)	1470 (28.2)	1.59	1.10
0.50[b]	87.5	17,700 (73.8)	1570 (26.2)	1.89	1.12
1.0	80.0	10,900 (65.2)	1620 (34.8)	1.60	1.20

[a]The value in parentheses is the molar percentage calculated from the GPC curve.
[b]Polymerized at 0°C.

The molecular weight distribution of the polymer was found to be dependent on the concentration of the monomer, [M]. As shown in Fig. 1 (Curve a) for the Asp.Bz system, only low molecular weight (M_w = 3400–5600) products with broad polydispersity (M_w/M_n = 3.50–5.11) were obtained when [Asp.Bz] was lower than 0.2 g/mL. On the other hand, when the [Asp.Bz] was greater than 0.2 g/mL, a polymer with a bimodal molecular weight distribution was observed [Fig. 1 (Curve b)]. Similar results were observed for the Glu.Bz system, i.e., when [Glu.Bz] = 0.5 g/mL, the highest M_w was obtained. The high molecular weight fraction constituted 60 to 75% of product for both systems. After a simple fractional precipitation from THF/methanol, poly(Asp.Bz) with M_w of 37,000 and M_w/M_n of 1.68 [Fig. 1 (Curve c)] was isolated in 45% yield from the polymer prepared with [Asp.Bz] = 1 g/mL. Similarly, poly(Glu.Bz) with M_w of 14,000 and M_w/M_n of 1.7 was also obtained in 50% yield.

Effect of Reaction Time and Temperature

A comparison between the polycondensations at room temperature (22°C) and at 0°C was carried out for the Glu.Bz system. Shown in Fig. 2 are the time-conversion curves for polymerization of Glu.Bz at room temperature and at 0°C. From this figure it can be seen that the polymerization at room temperature proceeded rapidly, achieving a yield of ~83% in 2 hours. Although the reactions at 0°C were slower, the final yields were similar.

A time-dependent study of the polymerization of Glu.Bz was carried out at room temperature. Shown in Fig. 3 are GPC curves that follow the progress of polymerization of Glu.Bz. After 15 minutes [Fig. 3 (Curve a)], most of the monomer still remained and only a small amount of polymer was detected. After 30 minutes [Fig. 3 (Curve b)], a significant amount of polymer was observed. Only a small amount of monomer remained in the reaction after 1 hour [Fig. 3 (Curve c)].

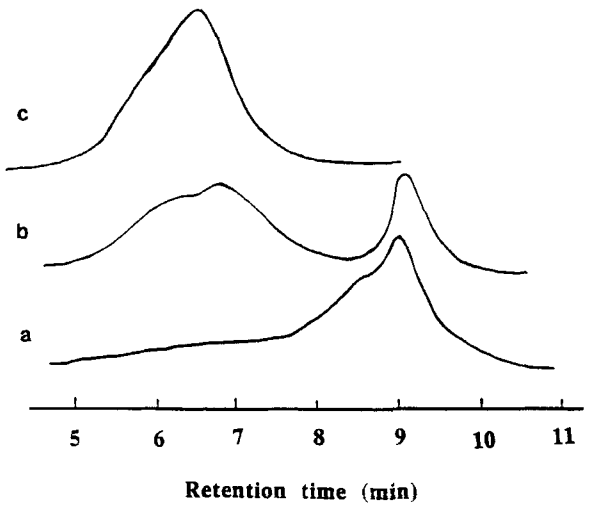


FIG. 1. GPC diagrams of poly(Asp.Bz) obtained from [Asp.Bz] = 0.025 g/mL (a), [Asp.Bz] = 1.0 g/mL (b), and [Asp.Bz] = 1.0 g/mL after fractionation (c).

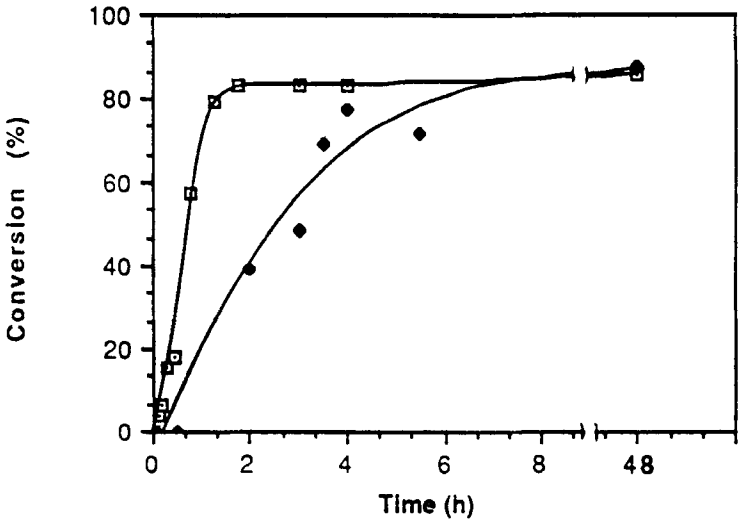


FIG. 2. Time-conversion curves for polycondensation of Glu.Bz in DMF at room temperature (□) and in DMF at 0°C (♦).

No significant change in molecular weight distribution appears to take place after 1 hour.

The relationship of molecular weight with reaction time is shown in Fig. 4 for Glu.Bz polymerized at room temperature and at 0°C. At room temperature, the molecular weight of the polymer increased rapidly with time during the first 3 hours and then remained constant. A maximum M_w of 17,000 (M_n = 10,900) was obtained. At 0°C, the molecular weight increased very slowly over a 48-hour period to provide similar M_w and M_n results.

The Asp.Bz polymerization ([Asp.Bz] = 1 g/mL) proceeded similarly, achieving 75% conversion in 4 hours and 85% in 6 hours at room temperature. Compared with Glu.Bz, the rate of Asp.Bz polymerization is considerably slower. The molecular weight of the polymer also increased during this 4 hours and then remained constant (Table 3). The high molecular weight fraction (M_w around 34,000) increased with time. A maximum content (76.8%) of the high molecular weight fraction was obtained after 43 hours.

The yield of poly(Asp.Bz) decreased with increasing reaction temperature. The polymerizations at 60 and 80°C gave only low molecular weight products (M_w = 860 and 640, respectively) in low yields (61.4 and 38.2%, respectively) (Table 3). The products were yellow colored and insoluble in THF but soluble in DMF and DMSO. The ^{1}H-NMR spectra of the products from the high temperature reactions were significantly different indicating more complicated structures. These may be due to side reactions that can occur at the high temperatures. It has been reported that a urea bond can be formed through a Curtius rearrangement during polycondensation by DPPA at higher temperature [47]. Consequently, the polymerization at room temperature seems to be the most favorable.

FIG. 3. GPC curves for poly(Glu.Bz) polymerization for 0.25 hour (a), 0.5 hour (b), 1 hour (c), and 24 hours (d) in DMF at room temperature.

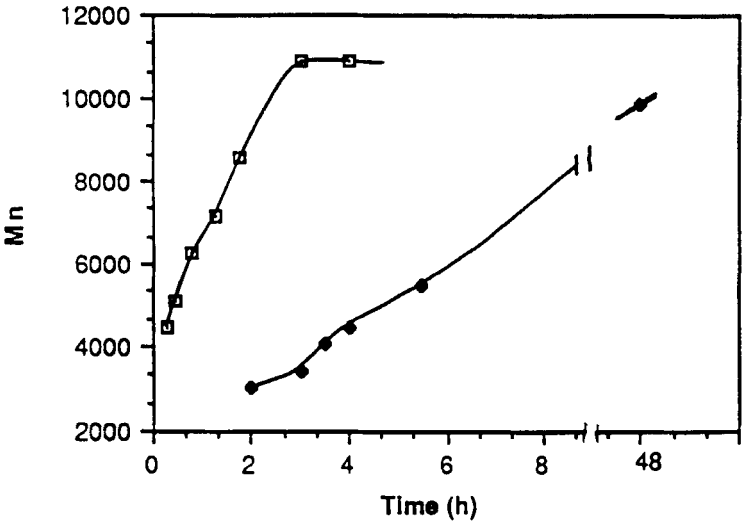


FIG. 4. Dependence of molecular weight of poly(Glu.Bz) on reaction time for the polycondensation in DMF at room temperature (□) and 0°C (♦).

Effect of Molar Ratios of [DPPA]/[Monomer] and [TEA]/[Monomer]

Nishi et al. reported that the polymer yield was dependent on the molar ratios of [DPPA]/[monomer] and [TEA]/[monomer] [46]. However, no data were reported with respect to the dependence of molecular weight or polydispersity on these ratios. The relationship between the molecular weight of the polymer and [DPPA]/[Asp.Bz], as well as [TEA]/[Asp.Bz], is shown in Table 4. The highest yield (86.5%) was obtained at a [DPPA]/[Asp.Bz] of 1.3 and a [TEA]/[Asp.Bz] of

TABLE 3. Effect of Reaction Time and Temperature on the Polymerization of Asp.Bz; [DPPA]/[M] = 1.3, [TEA]/[M] = 2.3, [M] = 0.5 g/mL

			GPC data			
Temperature, °C	Time, h	Yield, %	M_w,[a] Fraction 1	M_w,[a] Fraction 2	M_w/M_n, Fraction 1	M_w/M_n, Fraction 2
R.T.[b]	2	63.5	—	7900 (100)	—	5.65
R.T.[b]	4	75.3	34,000 (55.0)	1170 (45.0)	1.56	1.17
R.T.[b]	6	77.1	—[c]	—[c]	—[c]	—[c]
R.T.[b]	19	80.0	34,800 (63.5)	1030 (36.4)	1.64	1.18
R.T.[b]	43	83.5	34,000 (76.8)	1110 (23.2)	1.59	1.11
60	48	61.4	32,800 (1.2)	860 (98.8)	1.64	1.49
80	48	38.2	35,200 (3.2)	620 (96.8)	2.01	1.46

[a]The value in parentheses is the molar percentage calculated from the GPC curve.
[b]Polymerized at room temperature.
[c]Undetermined.

TABLE 4. Effect of the Molar Ratios of DPPA and TEA on Polymerization of Asp.Bz in DMF at Room Temperature for 2 days; [M] = 0.50 g/mL

[DPPA]/ [Asp.Bz]	[TEA]/ [Asp.Bz]	Yield, %	GPC data			
			M_w,[a] Fraction 1	M_w,[a] Fraction 2	M_w/M_n, Fraction 1	M_w/M_n, Fraction 2
0.5	2.3	16.3	—	3300 (100)	—	4.09
1.0	2.3	69.6	8,000 (45.4)	580 (54.6)	2.58	1.48
1.3	2.3	83.5	34,000 (76.8)	1100 (23.2)	1.59	1.11
1.5	2.3	69.4	28,100 (34.2)	1000 (65.8)	1.77	1.21
2.0	2.3	64.3	24,500 (58.3)	920 (41.7)	1.87	1.09
1.3	1.5	58.4	14,600 (39.3)	860 (60.7)	2.43	1.37
1.3	2.0	78.3	24,900 (64.3)	1100 (35.7)	1.87	1.19
1.3	3.0	74.6	26,900 (64.8)	980 (35.2)	1.98	1.18
1.3	3.5	65.0	22,400 (60.0)	880 (40.0)	2.70	1.10

[a]The value in parentheses is the molar percentage calculated from the GPC curve.

2.3. These observations are in agreement with the results reported by Nishi et al. [46] for the polymerization of other amino acids including Glu.Bz. It was found that the highest M_ws were also obtained with the highest yield at [DPPA]/[Asp.Bz] = 1.3 and [TEA]/[Asp.Bz] = 2.3, respectively (Table 4).

In the polycondensation by DPPA, for each mol of reacting Asp.Bz, 2 mol by-products, i.e., hydrazoic acid (HN_3) and diphenyl phosphoric acid, resulted. Therefore, at least 1 mol DPPA and 2 mol TEA are required. However, when excess amounts of DPPA (>1.3 mol) and TEA (>2.3 mol) were used, depolymerization might occur, which would lower the yield and molecular weight.

Therefore, it is clear that [DPPA]/[Asp.Bz] of 1.3 and [TEA]/[Asp.Bz] of 2.3 are the optimal conditions when both the yield and the molecular weight of the polymer are considered.

Effect of the Solvent

Because monomeric amino acid is insoluble in most organic solvents, the polymerization reaction was essentially a heterogeneous process in its early stages. The selection of solvents for polymerization was directed by the desire to dissolve the polymer product. The organic solvents used were DMF, DMSO, THF, acetonitrile, and dioxane as well as bulk polymerization. In all cases, the heterogeneous system became homogeneous after polymerization had progressed for 2 hours. A finely powdered monomer and effective stirring are important for efficient polymerization.

A comparison of the molecular weight of poly(Asp.Bz) prepared in different solvents, including bulk polymerization, is shown in Table 5. The molecular weight of the polymer was influenced by the solvents used. Although the yield was lower in THF (≃50%), a higher molecular weight polymer was obtained (M_w ≃ 38,800, M_w/M_n = 1.31). In contrast, polymerization in dioxane gave a lower molecular

TABLE 5. Effect of Solvents on the Polymerization of Asp.Bz at Room Temperature for 2 days; [DPPA]/[M] = 1.3, [TEA]/[M] = 2.3, [M] = 0.50 g/mL

Solvent	Yield, %	GPC data			
		M_w,[a] Fraction 1	M_w,[a] Fraction 2	M_w/M_n, Fraction 1	M_w/M_n, Fraction 2
DMF	86.5	24,400 (59.4)	990 (40.6)	2.22	1.08
DMSO	70.6	21,500 (78.9)	1190 (21.1)	1.87	1.13
THF	49.9	38,800 (74.6)	1290 (25.4)	1.31	1.13
Acetonitrile	71.1	33,500 (79.3)	1200 (20.7)	1.65	1.14
Dioxane	70.5	17,900 (68.5)	930 (31.5)	3.80	1.13
None[b]	61.2	39,300 (82.8)	1000 (17.2)	1.32	1.16

[a]The value in parentheses is the molar percentage calculated from the GPC curve.
[b]Bulk polymerization.

weight (M_w = 17,900) product with a broad polydispersity (M_w/M_n = 3.80). It is interesting to note that bulk polycondensation provided the highest M_w product (M_w = 39,300, DP = 200) in a yield of 61.2%.

Polymerization of Other Amino Acids

In order to compare DPPA polymerizations of other amino acids, L-phenylalanine (Phe), L-tyrosine (Tyr), L-Valine (Val), and L-isoleucine (Isoleu) were studied. The polycondensations were carried out in DMF {[monomer] = 0.5 g/mL} at room temperature using [DPPA]/[monomer] of 1.3 and [TEA]/[monomer] of 2.3. The results for these polymerizations are listed in Table 6 together with those for Asp.Bz and Glu.Bz.

TABLE 6. Comparison of Polycondensation of Various Amino Acids[a]

Amino acid	Yield, %	M_w,[b] GPC
Asp.Bz	86.5	25,200 (65.4)
Glu.Bz	85.7	24,400 (71.8)
Phe	95.0	15,100 (23.3)
Tyr	85.0	14,300 (42.1)
Val	Trace	–
Isoleu	Trace	–

[a]Polymerized in DMF at room temperature: [DPPA]/[monomer] = 1.3; [TEA]/[monomer] = 2.3; [monomer] = 0.5 g/mL.
[b]For higher molecular weight fraction. Determined using μStyragel column (10^3 Å, Waters, Inc.) in 0.1% LiCL/DMF at 70°C (polystyrene standards). The value in parentheses is the molar percentage calculated from the GPC curve.

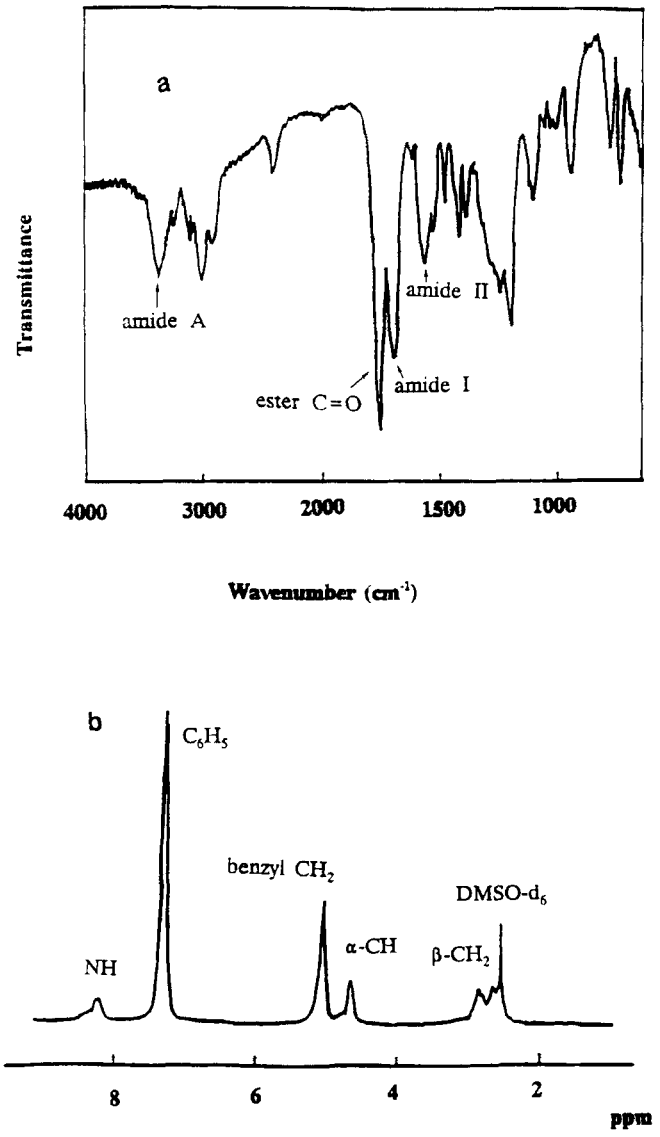


FIG. 5. IR (a) and ^{1}H-NMR (b) spectra of poly(Asp.Bz).

For Phe and Tyr systems, polymerizations gave polymers in good yields (>85%). Bi- or multimodel molecular weight distributions were observed in both cases. Phe and Tyr gave lower molecular weight products (M_w = 15,100 and 14,300, respectively) compared with Asp.Bz and Glu.Bz. The contents of the high molecular weight fractions were also lower (23.3 and 42.1%, respectively). These may partially be due to their poorer solubilities in DMF. However, there is still the possibility that the reactivity is influenced by the electric and steric factors of the sidechain group. This is supported by the fact that Val and Isoleu, which have a bulky sidechain group, did not undergo polycondensation under the present conditions (Table 3).

Polymerizations of L-, D-, and DL-amino acids by DPPA gave similar molecular weights and polydispersities. A more complete study of these systems is being conducted.

Characterization of the Polymer

The structures of these polymers were studied by IR and ^{1}H-NMR spectroscopy. The IR and ^{1}H-NMR spectra of poly(L-Asp.Bz) are shown in Fig. 5. From the IR spectrum, the characteristic peptide absorb bands were observed at 3330 (amide A), 1672 cm^{-1} (amide I), and 1543 cm^{-1} (amide II), respectively, as well as at 1735 cm^{-1} for ester C=O. From the ^{1}H-NMR spectrum, the characteristic peaks of the amino acid repeat unit, i.e., NH at 8.20 ppm, C_6H_5 at 7.2 ppm, benzyl CH_2 at 4.95 ppm, α-CH at 4.65 ppm, and β-CH_2 at 2.70 ppm, were clearly observed. Based on the spectral data, the polymers prepared through direct polycondensation by DPPA have the same structure as those obtained by the NCA method.

CONCLUSION

1. Poly(amino acid)s with molecular weights up to 40,000 for poly(Asp.Bz)s and 20,000 for poly(Glu.Bz)s were prepared by the direct polycondensation of amino acids using DPPA as a condensation agent. Low molecular weight products were obtained for Phe (4,300) and Tyr (14,300) while Val and Isoleu did not produce a polymer.

2. In most cases, a bimodal molecular weight distribution was observed.

3. The yield of the high molecular weight fraction was dependent on the monomer concentration, reaction time, and temperature.

4. Maximum molecular weight and yield of polymer were obtained at ratios of [DPPA]/[monomer] = 1.3 and [TEA]/[monomer] = 2.3.

5. The molecular weights of the poly(amino acid)s are influenced by the solvents used, while bulk polymerization gave the highest molecular weight.

ACKNOWLEDGMENTS

The authors wish to thank Emisphere Technologies, Inc., for financial support. Grateful acknowledgment is also due to S. I. Zaidi for GPC and NMR measurements, and to Dr. S. A. George and J. L. Ding for technical assistance.

REFERENCES

[1] J. Kohn, *Biodegradable Polymers as Drug Delivery Systems* (M. Chasin and R. Langer, Eds.), Dekker, New York, 1990.

[2] J. M. Anderson, K. L. Spilizewski, and A. Hiltner, *Biocompatibility of Tissue Analogs*, Vol. 1 (D. F. Williams, Ed.), CRC Press, Boca Raton, Florida, 1985.

[3] H. Leuchs, *Ber. Dtsch. Chem. Ges., 39*, 859 (1906).

[4] H. Leuchs and W. Mannasse, *Ibid., 40*, 3235 (1907).

[5] H. Leuchs and W. Geiger, *Ibid., 41*, 1721 (1908).

[6] H. Block, *Poly(γ-Benzyl-L-Glutamate) and Other Glutamic Acid Containing Polymers*, Gordon and Breach, New York, 1983.
[7] J. W. Stewart and M. A. Stahmann *Polyamino Acids, Polypeptides and Proteins*, Vol. 15 (M. A. Stahmann, Ed.), University of Wisconsin Press, Madison, 1962.
[8] C. H. Bamford and H. Block, *The Polymerization of N-Carboxy-α-Amino Acids* (C. H. Bamford and C. F. H. Tipper, Eds.), Elsevier, Amsterdam, 1976.
[9] J. Miquel, S. Brooke, and S. W. Fox, *Curr. Mod. Biol., 3*, 299 (1971).
[10] L. L. Hsu, S. Brooke, and S. W. Fox, *Ibid., 4*, 12 (1971).
[11] J. W. Ryan and S. W. Fox, *BioSystems, 5*, 115 (1973).
[12] S. W. Fox, *Naturwissenschaften, 67*, 378 (1980).
[13] J. C. Lacey Jr., A. Yuki, and S. W. Fox, *BioSystems, 11*, 1 (1979).
[14] G. Vaughan, A. T. Przybylski, and S. W. Fox, *Ibid., 20*, 219 (1987).
[15] W. W. McAlhaney and D. L. Rohlfing, *Ibid., 8*, 45, (1976).
[16] W. D. Snyder and S. W. Fox, *Ibid., 7*, 222 (1975).
[17] R. M. Syren, A. Sanjur, and S. W. Fox, *Ibid., 17*, 275 (1985).
[18] S. W. Fox, J. R. Jungck, and T. Nakajima, *Origins Life, 5*, 227 (1974).
[19] L. Hsu and S. W. Fox, *BioSystems, 8*, 89 (1976).
[20] S. Brooke and S. W. Fox, *Ibid., 9*, 1, (1977).
[21] D. L. Rohlfing, *Origins Life, 6*, 203 (1975).
[22] S. W. Fox, *Ibid., 7*, 49 (1976).
[23] E. Kokufuta, H. Sakai, and K. Harada, *BioSystems, 16*, 175 (1984).
[24] S. Steiner and R. Rosen, U.S. Patent 4,925,673 (1990).
[25] M. Bodanszky, *Principles of Peptide Synthesis*, Springer-Verlag, Berlin, 1984.
[26] M. Bodansky and A. Bodansky, *The Practice of Peptide Synthesis*, Springer-Verlag, Berlin, 1984.
[27] M. J. Fournier, H. S. Creel, M. T. Krejchi, T. L. Mason, D. A. Tirrell, K. P. McGrath, and E. D. T. Atkins, *J. Bioact. Compat. Polym., 6*, 326 (1991).
[28] N. Ogata, K. Sanui, and S. Tan, *Polym. J. (Tokyo), 16*, 569 (1984).
[29] H. Tanaka, G. C. Wu, Y. Iwanaga, K. Sanui, and N. Ogata, *Ibid., 14*, 331 (1982).
[30] G. C. Wu, H. Tanaka, K. Sanui, and N. Ogata, *Ibid., 14*, 797 (1982).
[31] G. C. Wu, H. Tanaka, K. Sanui, and N. Ogata, *Ibid., 14*, 571 (1982).
[32] H. Tanaka, G. C. Wu, Y. Iwanaga, K. Sanui, and N. Ogata, *Ibid., 14*, 635 (1982).
[33] K. Sanui and N. Ogata, *Proc. IUPAC, Macromol. Symp., 28th*, p. 120 (1982).
[34] F. Higashi, S. Ogata, and Y. Aoki, *J. Polym. Sci., Polym. Chem. Ed., 20*, 2081 (1982).
[35] F. Higashi, Ger. Offen. DE 3,316,137, Cl. C08G63/38 (1984).
[36] F. Higashi, N. Akiyama, I. Takahashi, and T. Koyama, *J. Polym. Sci., Polym. Chem. Ed., 22*, 1653 (1984).
[37] F. Higashi and T. Mashimo, *J. Polym. Sci., Part A: Polym. Chem., 24*, 1697 (1986).
[38] F. Higashi, A. Hoshio, and J. Kiyoshige, *Makromol. Chem., Rapid Commun., 4*, 371 (1983).
[39] F. Higashi, *Yuki Gosei Kagaku Kyokaishi, 40*, 922 (1982).
[40] F. Higashi, *Kobunshi, 32*, 438 (1983).
[41] T. Shioiri and S. Yamada, *Chem. Pharm. Bull., 22*, 855 (1974).

[42] T. Shioiri and S. Yamada, *Ibid., 22*, 859 (1974).
[43] T. Shioiri, K. Ninomiya, and S. Yamada, *J. Am. Chem. Soc., 94*, 6203 (1972).
[44] N. Nishi, B. Nakajima, N. Hasebe, and J. Noguchi, *Int. J. Biol. Makromol., 2*, 53 (1980).
[45] B. Nakajima and N. Nishi, *Polym. J. (Tokyo), 13*, 183 (1981).
[46] N. Nishi, M. Tsunemi, H. Hayasaka, B. Nakajima, and S. Tokura, *Makromol. Chem., 192*, 1789 (1991).
[47] N. Nishi, T. Naruse, K. Hagiwara, B. Nakajima, and S. Tokura, *Ibid., 192*, 1799 (1991).
[48] N. Nishi, M. Tsunemi, K. Nakamura, and S. Tokura, *Ibid., 192*, 1811 (1991).

Received December 11, 1992.