JOURNAL OF MATERIALS SCIENCE **38** (2003) 3191–3197

Photoresponsive peptide and polypeptide systems 15*: Synthesis of photo-crosslinkable poly(amino acid)s by watery process and its application as a reinforcement for polyion complex fibers

K. OHKAWA[‡], K. SHOUMURA, Y. SHIRAKABE, H. YAMAMOTO
Institute of High Polymer Research, Faculty of Textile Science and Technology,
Shinshu University, Ueda 386-8567, Japan
E-mail: kohkawa@giptc.shinshu-u.ac.jp

The present article described the synthesis of the coumarin-containing polypeptides using the water-soluble active ester method and the reinforcement of the polyion complex (PIC) fibers by the photochemical cross-linking between the polypeptide-pendant coumaryloxyacetyl groups. Two kinds of cationic polypeptides, poly(α,L-lysine) (PLL) and poly(α,L-ornithine) (PLO) were treated with (4-hydroxyphenyl)dimethyl sulfonium methyl sulfate ester in water to incorporate the photoreactive coumaryloxyacetyl groups into the side chain amino groups of the PLL and PLO. The contents of the photoreactive groups in the produced coumarin-containing polypepides were controlled by the molar ratios of the amino groups to the active ester. The resulting coumarin-containing cationic polypeptides were spun into the PIC fibers with the anionic polysaccharide gellan in water. The tensile strengths of the coumarin-containing PIC fibers were superior to those of the intact PLL- and PLO-gellan PIC fibers. The mechanical strength of the coumarin-containing PIC fibers further increased upon light-irradiation. From these results, we concluded that the hydrophobic interactions and the covalent cross-linking due to the photochemical dimerization between the coumaryloxyacetyl groups significantly contributed to reinforcement of the PIC fibers. The present results afford a new methodology to the PIC reinforcement that was achieved by the polymer modification reaction in watery process.

1. Introduction

The crosslinking reaction of polymeric materials leads to mechanical reinforcement, insolubilization, and resistance to chemical degradation [2, 3]. Thus, there are many applications of cross-linking methods for material formulations as fibers, films, gels, capsules, and adhesives from polymer solutions [4]. The formation of dimers and cycloaddition products upon light-irradiation of compounds containing olefinic constituents was one of the first types of photochemical reactions but it is still a useful strategy for the formation of a cross-linking polymer network [5].

Photosensitive biopolymers are interesting systems because of their relevance to the molecular mechanism of photochemistry in biological materials and processes and have received increased attention due to their broad applications as new formulation systems. Our group has been studying the photoresponsive peptide and polypeptide systems [1, 6]. The major objective in this research field is the design of a biopolymer system reversibly responsible for photo-induced events, including new methods for the synthesis of photoresponsive polymers, the dimerization mechanism of the side chain photoresponsive groups, and the material science associated with the cross-linked materials [7]. When a photosensitive group is introduced into the polypeptide side chains, the photodimerization upon irradiation causes interchain crosslinking, resulting in the insolubilization or gelation of the polymer. Cinnamoyl [8], thymine [9], and coumarin [6] have been utilized as the polypeptide-bound photoresponsive groups capable of inducing the photo-cross-linking.

Recently, we found the characteristic interfacial structures, such as films, fibers, and capsules, which are created from the electrostatic polyion complex (PIC) formation between a polyelectrolyte and an oppositely

---

*Part 14: cf. reference [1].

[‡] Author to whom all correspondence should be addressed.

0022–2461

charged polyelectrolyte in aqueous solution [10]. Considering that almost all biopolymers are polyelectrolytes, the study of PIC as models of complicated biological systems and as material formulations are very important [11]. A fabrication method via PIC formation is a unique strategy that realizes the formulation of poly(amino acid) fibers from their aqueous solutions. Our attempt at the development of new types of poly(amino acid) PIC fibers is a kind of reactive spinning [4]. We have reported the spinning methods and characterizations of five kinds of PIC fibers, i.e., chitosan-gellan [11], poly(α,L-glutamic acid)-chitosan [12], poly(α,L-lysine)-gellan [13], poly(α,L-glutamic acid)-poly(α,L-lysine) [14], and chitosan-poly(acrylic acid) [15] fibers. These five PIC fibers can be prepared by self-assembling at the aqueous solution interface.

The tensile strength of the PIC fibers is enhanced by the cross-linking reaction using several kinds of bifunctional cross-linking reagents [11, 13]. In the present study, we attempted the reinforcement of the mechanical strength of the PIC fibers using a photo-induced cross-linking reaction. As the photoreactive group for the photo-induced dimerization, coumarin [16–18] was chosen because of its usefulness in our previous studies for preparing photo-cross-linking materials [1, 6]. In our previous studies, coumarin-containing cationic polypeptides, copoly[LysLys(Cou)] and copoly[OrnOrn(Cou)], were prepared by the copolymerization reactions of the N-carboxylic anhydrides of N^{ε}-Z-lysine [Lys(Z)NCA] and N^{ε}-coumaryloxyacetyl-lysine [Lys(Cou)NCA] and of the corresponding derivatives of ornithine [Orn(Z)NCA and Orn(Cou)NCA], followed by deprotection of the Z groups. The copolymerization method itself has the advantage that the content of the coumaryloxyacetyl groups is controlled by the monomer feed ratio. The series of copolymerized samples having various coumaryoxyacetyl group contents, however, have the molecular weight diversity [1, 6].

The mechanical properties of the fibers generally depend on the molecular weight of the parent polymer. In order to discuss the effect of the photo-cross-linking reaction on the mechanical reinforcement of the PIC fibers, the effect of the molecular weight of the parent polymer on the mechanical strength of the PIC fiber should be eliminated. Hence, in the present study, we employed the water-soluble active ester method to introduce the coumaryloxyacetyl group into the amino groups of poly(α,L-lysine) and poly(α,L-ornithine). This method allows the forming of an amide bond under aqueous conditions, and the resulting modified polymer has the same molecular weight as the parent one. The ester of (4-hydroxyphenyl)dimethylsulfonium methyl sulfate (HOPDS) has been developed as a water-soluble active ester, which is suitable for the purpose of the chemical modification of proteins without denaturation [19]. The HOPDS method was chosen to synthesize the coumarin-containing polypeptide due to the water solubility of the cationic polypeptides. We describe here the synthesis of the coumarin-containing polypeptide by the water-soluble active ester method in order to spin the PIC fiber, and reinforcement of the fiber by photochemical cross-linking between the coumaryloxyacetyl groups.

2. Experimental procedures

2.1. Materials

Poly(α,L-lysine) (PLL) and poly(α,L-ornithine) (PLO) was prepared according to the modified method as described in our earlier article [14]. The viscosity average molecular weights of PLL hydrobromide and PLO hydrobromide are, respectively, 385,000 [degree of polymerization (DP) is 1840] and 111,000 (DP 570) from the viscosity equation $\log \mathrm{DP} = 1.47 \times \log[\eta] + 2.99$ for the protected poly(N^{ε}-benzyloxycarbonyl-L-lysine) [Lys(Z)] [20]. Gellan gum [molecular weight (MW) 880,000–920,000] was from San-Ei-Gen F.F.I., Inc. (4-Hydroxyphenyl)dimethylsulfonium methyl sulfate (HOPDS) was purchased from Sanshin Chemical Industrial Co., Ltd. Ethylbromoacetate, triethylamine (TEA), dicyclohexylcarbodiimide (DCCI) and N,N-dimethylformamide (DMF) were purchased from Wako Pure Chemical Industries, Ltd. 7-Hydroxycoumarin was purchased from Tokyo Chemical Industry Co., Ltd. Coumaryloxyacetic acid (Cou-OH) was prepared as previously described [6].

2.2. Coumaryloxyacetic acid (4-hydroxyphenyl)dimethyl sulfonium methyl sulfate ester (Cou-OPDS)

Cou-OH (14.9 g, 67.7 mmol) and HOPDS (18.0 g, 81.1 mmol) were dissolved in DMF (6×10^{-5} m^3), and DCCI (16.7 g, 80.9 mmol) was added to the solution at room temperature with stirring. After stirring for 3 h at 40°C, dicyclohexylurea was filtered off, and diethyl ether (5×10^{-4} m^3) was added to the filtrate. The crude product obtained as white powder which was recrystallized from DMF and diethyl ether. Yield 24.7 g (78%); m.p. = 125–127°C; thin layer chromatography 95:5:3 chloroform/methanol/acetic acid, $R_{\mathrm{f}} = 0.66$; analysis for $C_{20}H_{20}O_9S_2$: Calc. C 51.27, H 4.30; Found. C, 51.10, H 4.31.

2.3. Synthesis of coumarin-containing polypeptides

A typical experimental procedure is as follows. Cou-OPDS (0.22 g, 0.48 mmol) was added to a solution of PLL (1.00 g, 4.78 mmol) in H_2O (1.5×10^{-5} m^3). To the soulution was immediately added 1 M TEA in dioxane (4.8×10^{-7} m^3). After stirring for 4 h at 40°C, the solution was dialyzed against H_2O. A small amount of the insoluble material was removed by filtration. The produced coumarin-containing PLLs were obtained as aqueous solutions. The yields were estimated as the weights of the dissolved polymer per 2×10^{-6} m^3 of the product solutions (90–95%). The coumarin-containing PLO samples were prepared using the same procedure.

2.4. Preparation of polyion complex (PIC) fiber

For the PIC fiber spinning, the polypeptide solutions were prepared at the concentration of 5.0%, and the pH values were adjusted using 0.1 M HCl. Gellan gum was dissolved in hot water at a concentration of 0.75%. The PIC fibers were prepared using a simple wet-spinning apparatus accompanied by a roller as reported in our earlier articles [11, 15]. In brief, the 5.0% coumarin-containing polypeptide solution was slowly added onto a 0.75% gellan solution at 60°C. Immediately the polypeptides and gellan spontaneously associated to form the PIC film at the interface between the polymer solutions, and the PIC film was drawn by pincettes to spin a wet thread at the rate of 20 mm $\cdot$ s^{-1}. The formed thread was passed through an ethanol bath (length, 1.5 m; depth, 35 mm) and rolled up at the rate of 1.2 m $\cdot$ min^{-1} with drying using hot air.

2.5. Evaluation of mechanical properties of PIC fibers

The mechanical properties of the PIC fibers were evaluated using a tensile testing machine (PS-5K, Imada Co.). A looped test thread was set on the testing machine equipped with a hook jig. The load was generated at 10 mm $\cdot$ min^{-1} and the breaking load value (N) was recorded. The tensile strengths per one PIC fiber (MPa) were then calculated from the average breaking load value (gf) of ten independent measurements and the cross-sectional area A (mm^2) was estimated from the equation, $A = (\phi/2)^2 \cdot \pi$, where ϕ is the average diameter (mm) of the fiber observed under a microscope.

The stress-strain curves upon uniaxial drawing of a single thread of the PIC fiber were measured using a Tensilon (STA-1150, Orientec Co.). The initial gauge length was 20 mm, and the drawing speed was 20 mm $\cdot$ min^{-1}. The average stress-strain curves were determined from 20 independent measurements. The breaking stress (MPa) was defined as the load (N) per A (mm^2) value at the rupture point of a specimen. The strain (%) was calculated from the increase in length of a specimen during a tensile test, expressed as a percentage of the initial gauge length.

2.6. Photochemical reactions for fiber reinforcements

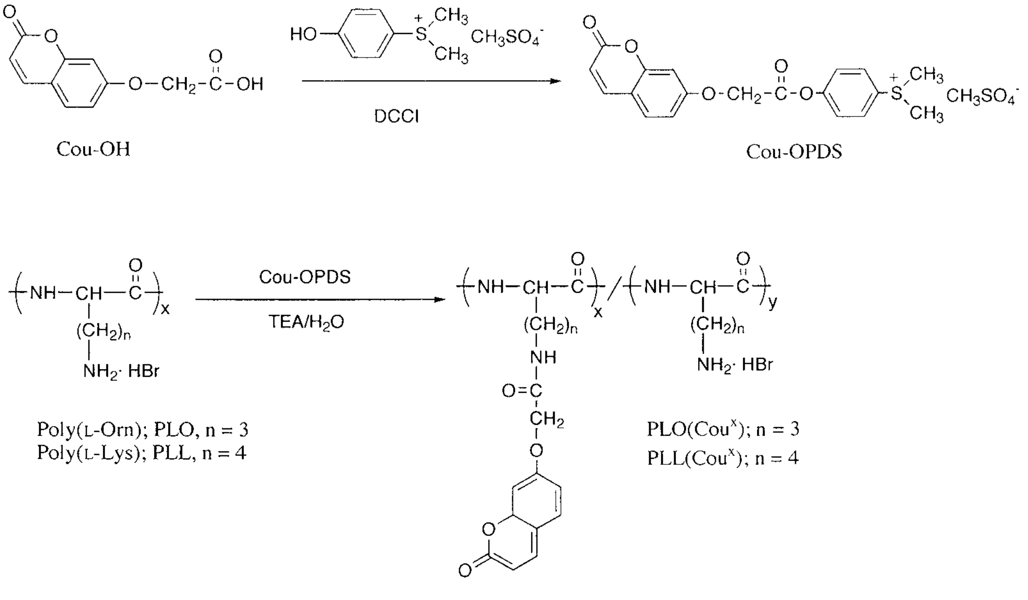
Irradiations of the PIC fibers were carried out with a high-pressure mercury lamp (400 W) without a filter at room temperature. The light intensity was determined by chemical actinometry using potassium ferrioxalate to be 5.1×10^{16} photons $\cdot$ $mm^{-2} \cdot s^{-1}$ [6].

3. Results and discussion

3.1. Synthesis of coumarin-containing polypeptide using water-soluble active esters

According to Scheme 1, the coupling reaction between the side chain amino groups of PLL/PLO and the activated carbonyl group of the Cou-OPDS was carried out in a water solution. The feed molar ratio of Cou-OPDS towards the amino group was changed in order to obtain coumarin-containing PLL and PLO having various coumaryloxyacetyl side chain contents. The degree of substitution (DS) was determined from the molecular extinction coefficient of Lys(Cou). The DS values of the coumarin-containing PLL and PLO are listed in Table I. The DS values were roughly controlled by the feed mole ratio of Cou-OPDS to the side chain amino groups of PLL and PLO.

Since the substituted side chain coumaryloxyactectyl group is hydrophobic, the coumarin-containing PLL having the DS value above 0.15 was insoluble in water. Previously, we reported the synthesis of the coumarin-containing PLL by copolymerizing Lys(Z)NCA and Lys(Cou)NCA followed by deprotection of the Z group, and a similar synthetic procedure using Orn(Z)NCA and Orn(Cou)NCA procuced the coumarin-containing PLO. The coumarin-containing PLL and PLO obtained by the copolymerization method were soluble in water up to the DS values of approximately 0.33 [1, 6]. The differences in the water solubility between our present and previous samples are probably due to the difference in their molecular weights. In fact, the present samples have much higher DP values (PLL, DP = 1840; PLO,



Scheme 1

TABLE I Synthesis of PLL(Cou^{x}) and PLO(Cou^{x}) by the water-soluble active ester method

Parent polymer	Equivalent molar of Cou-OPDS	DS (%)[a]	Yielded polymer
PLL[b]	0.10	0.054	PLL(Cou^{5})
	0.20	0.10	PLL(Cou^{10})
	0.35	0.14	PLL(Cou^{14})
PLO[c]	0.10	0.05	PLO(Cou^{5})
	0.20	0.10	PLO(Cou^{10})
	0.50	0.22	PLO(Cou^{22})

[a] Estimated from the molecular extinction coefficient of the monomeric L-Lys(Cou) or L-Orn(Cou) in water.
[b] Poly(L-lysine).
[c] Poly(L-ornithine).

DP = 570) than the previous ones (PLL, DP = 110–310; PLO, DP = 150–180) and thus are less soluble in water at their higher DS values.

3.2. Mechanical reinforcement by photo-crosslinking reaction

3.2.1. Loop thread test

The PLL(Cou^{x})-gellan and PLO(Cou^{x})-gellan PIC fibers possess a silk-like luster. Long monofilament fibers can be obtained in both cases, and as an example, Fig. 1 shows the rolled-up and dried PLL(Cou^{14})-gellan PIC fiber.

At first, the tensile strength of the PIC fiber samples was evaluated by the loop thread test to investigate the optimal condition for the photochemical reinforcement. Fig. 2 shows the relationship between the fiber tensile strength and the irradiation time. The tensile test was performed using the looped thread as described under the experimental procedure section. Before irradiation (0 min), the tensile strength of the PLL(Cou^{x})-gellan fibers increased in the following order; PLL (170 MPa) < PLL(Cou^{5}) (207 MPa) < PLL(Cou^{10}) (234 MPa) < PLL(Cou^{14}) (284 MPa). This result indicated that the fibers with a higher coumarin content exhibited a higher tensile strength. A similar increase in the tensile strength was found in the series of PLO(Cou^{x})-gellan and the intact PLO-gellan PIC fibers (Fig. 3).

As the irradiation time increased from 0 to 30 min, the tensile strengths in the PLL(Cou^{x})-gellan and PLO(Cou^{x})-gellan PIC fibers increased. The greatest effect to enhance the tensile strength of the PIC fibers was obtained when the copoly PLL(Cou^{14})-gellan fiber was irradiated. The initial strength of the PLL(Cou^{14})-gellan fiber (284 MPa) increased to 486 MPa after a 30-min irradiation (Fig. 2). This result indicated that a 1.7-fold reinforcement of the PIC fiber was achieved by the photo-induced crosslinking reaction of PLL(Cou^{14}). As compared to the intact PLL-gellan fiber after a 30-min irradiation (163 MPa), the photo-crosslinked PLL(Cou^{14})-gellan fiber exhibited a 3-fold higher tensile strength. The tensile strengths of the photo-cross-linked PLL(Cou^{x})-gellan fibers after a 30-min irradiation were in the following order; PLL(Cou^{14}) (486 MPa) > PLL(Cou^{10}) (349 MPa) > PLL(Cou^{5}) (270 MPa). These results indicate that the effect of the photo-induced reinforcement of the PLL(Cou^{x})-gellan PIC fibers depend on their coumaryloxyacetyl groups.

The photo-induced reinforcement was observed in a similar manner for the PLO(Cou^{x})-gellan fibers. The tensile strength of the photo-cross-linked PLO(Cou^{22}) after a 30-min irradiation was 376 MPa, and this value was 1.3-fold higher than that of the non-irradiated sample (279 MPa). As compared with the intact PLO-gellan fiber after a 30-min irradiation (195 MPa), the photo-cross-linked PLL(Cou^{14}) fiber exhibited a 1.9-fold higher strength. When the irradiation time was longer

Figure 1 Photograph of PLL(Cou^{14})-gellan PIC fiber.

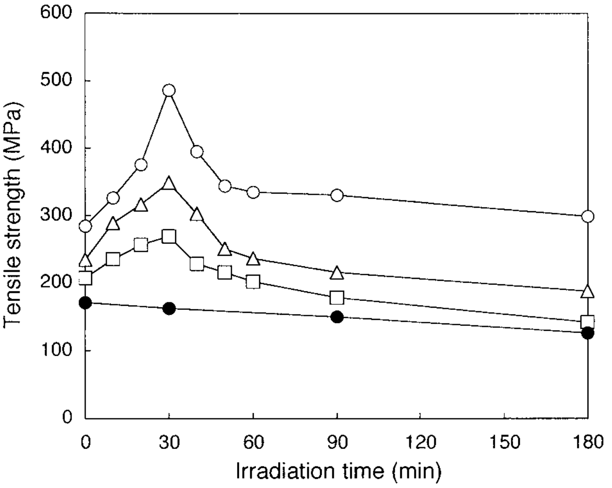


Figure 2 Relationship between the irradiation time and the tensile strength of photo-cross-linking PLL(Cou^x)-gellan PIC fibers: (○) PLL(Cou^{14}); (△) PLL(Cou^{10}); (□) PLL(Cou^5); (●) PLL.

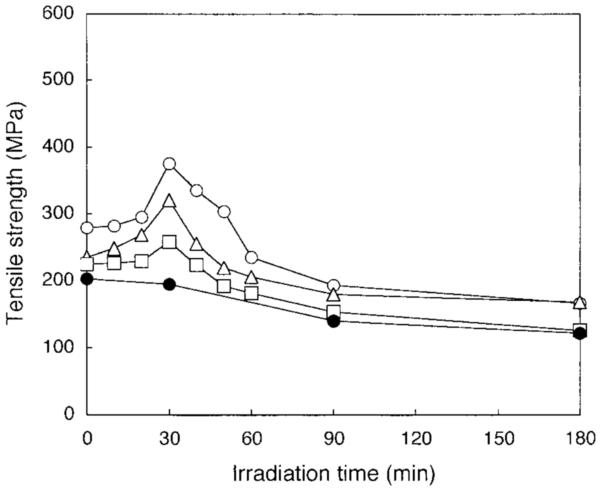


Figure 3 Relationship between the irradiation time and the tensile strength of photo-cross-linking PLO(Cou^x)-gellan PIC fibers: (○) PLO(Cou^{22}); (△) PLO(Cou^{10}); (□) PLO(Cou^5); (●) PLO.

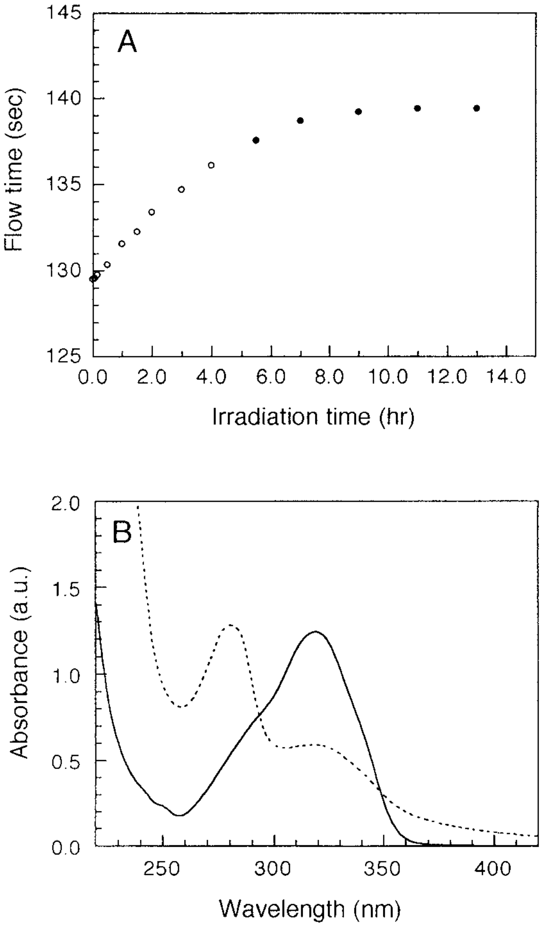


Figure 4 Panel A, the flow time changes in viscometry upon the UV-irradiation on the PLO(Cou^{10}) aqueous solution; (●) indicates the apparent flow time data after the insolubilization occurred. Panel B, absorption spectra of PLO(Cou^{10}) solutions after the UV-irradiation for 0 h (solid line) and 20 h (broken line).

than 30 min, the tensile strength of the PLL(Cou^x)-gellan and PLO(Cou^x)-gellan fibers decreased. Therefore, the optimal irradiation time for the maximum reinforcement was found to be 30 min.

As one possibility for the decrease in the tensile strength, the photochemical cleavage of the polyamide backbone is considered [21, 22]. To discuss the molecular mechanism on the decrease of the tensile strength of the PLL(Cou^x)-gellan and PLO(Cou^x)-gellan PIC fibers, the photochemical degradation of PLO(Cou^{10}) was examined. In our present study, once the PIC fiber is formed, the parent PLO(Cou^x) or PLL(Cou^x) cannot be solubilized from the PIC fiber matrix, otherwise chemically destructive methods such as acid hydrolysis. Thus, the photo-induced viscosity change was measured upon the UV-irradiation on the PLO(Cou^{10}) aqueous solution.

Fig. 4A represents the change in the flow time of the PLO(Cou^{10}) aqueous solution (5%) in the viscometry upon the UV-irradiation. The flow time increased during the first 5 h (Fig. 4A, open circles). This result indicates that the intermolecular photochemical cross-linking reaction proceeds to increase the molecular weight of the PLO(Cou^x) samples. After a 5-h irradiation, the cross-linked polymer precipitates produced in the viscometer, interfering with the accurate flow time measurement. The apparent flow times (Fig. 4A, closed circles) reached to plateau at 11 h. For irradiation exposures longer than 11 h, the PLO(Cou^{10}) solution gradually turned dark brownish, implying the photo-induced oxidative damage in the PLO(Cou^{10}) molecules.

Next, the gel permeation chromatography (GPC) analysis was performed to examine the molecular weight distribution of the PLO(Cou^{10}) upon the UV-irradiation. The 20-h irradiated PLO(Cou^{10}) aqueous solution was filtered to remove the cross-linked precipitates, and the filtrate was analyzed using a Shodex KW-803 GPC column (Showa Denko, Co. LTD., Japan). The molecular weight distribution of the irradiated PLO(Cou^{10}) was almost identical to that of the non-irradiated sample, indicating that the photochemical degradation did not occur at the PLO polyamide backbone to decrease the molecular weight (data not shown).

When the UV-spectrum of the 20-h irradiated PLO(Cou^{10}) solution compared to that of the non-irradiated sample, a clear difference was observed (Fig. 4B). We have previously reported the differences in UV-spectral changes of the monomeric Lys(Cou) and

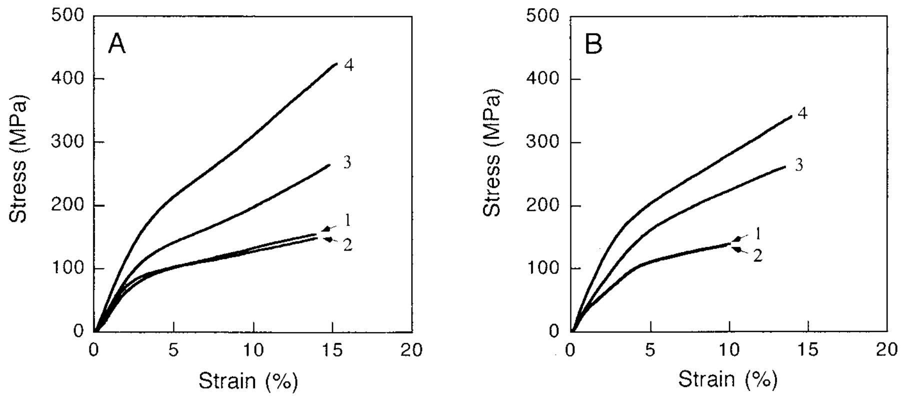


Figure 5 Stress-strain curves of photo-cross-linking PIC fibers. Panel A, PLL-gellan PIC fibers (curve 1, before irradiation; curve 2, after a 30-min irradiation) and PLL(Cou^{14})-gellan PIC fibers (curve 3, before irradiation; curve 4; after a 30-min irradiation). Panel B, PLO-gellan PIC fiber (curve 1, before irradiation; curve 2, after a 30-min irradiation) and PLL(Cou^{22})-gellan PIC fibers (curve 3, before irradiation; curve 4; after a 30-min irradiation).

of the coumarin in their photochemical dimerization reaction [6]. The present spectrum of the 20-h irradiated PLO(Cou^{10}) solution is characteristic for the monomeric coumarin (λ_{max}, 273 nm), but not for the monomeric Lys(Cou) (λ_{max}, 228 nm). These results suggest that the photochemical degradation might occur at the cross-bridge structures (dimerized coumaryloxyacetyl side chains) to cleave the cross-linkings and finally eliminate the coumaryloxyacetyl moieties from the PLO(Cou^{x}) molecules.

3.2.2. Uniaxially drawing test

Finally, we evaluated the mechanical properties of the coumarin-containing PIC fibers by the uniaxially drawing test. The load values in N were normalized for a comparison per cross-sectional area (mm^2). Fig. 5A shows the average stress-strain curves of the PLL(Cou^{14})-gellan and the intact PLL-gellan fibers. The irradiation time was fixed at 30 min due to the results in Figs 2 and 3. The breaking load (MPa) of the intact PLL-gellan fibers was 155 MPa at the 14% strain (Fig. 5A, curve 1). The irradiated control sample (Fig. 5A, curve 2) exhibited almost the same stress-strain curve as that of the intact sample.

As for the PLL(Cou^{14})-gellan fiber, the breaking stress of the non-irradiated sample was 265 MPa (Fig. 5A, curve 3), and this value was 1.7-fold higher than that of the intact PLL-gellan fiber (Fig. 5A, curve 1). Likewise in the case of the loop thread test, the effect of the incorporation of the coumaryloxyacetyl side chains on the increase of tensile strength of the PIC fiber was found in the present uniaxally drawing test. The strain at rupture of the PLL(Cou^{14})-gellan fiber (15%) was slightly higher than that of the intact PLL-gellan fiber (14%).

When the PLL(Cou^{14})-gellan fiber was irradiated for 30 min, the breaking stress significantly increased (425 MPa, Fig. 5A, curve 4) as compared to the non-irradiated sample (Fig. 3A, curve 3). The 1.6-fold increase upon irradiation in the uniaxially drawing test was comparable to that of the loop thread test (1.7 fold, Fig. 2).

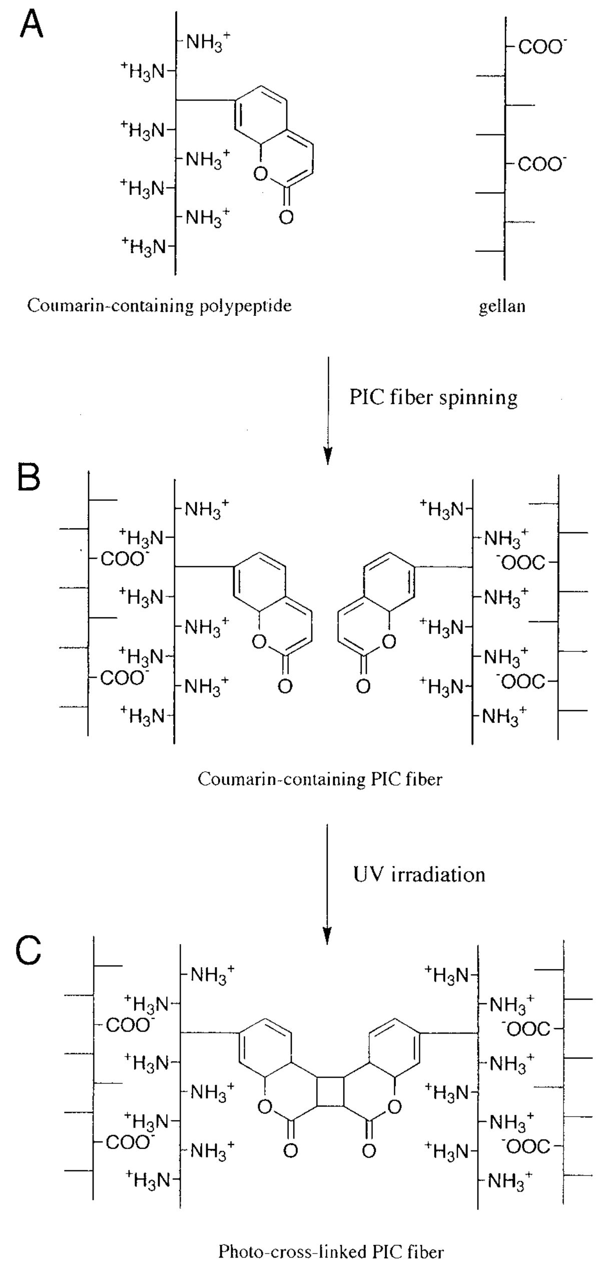
Fig. 5B represents the results of similar experiments using the intact PLO-gellan and PLO(Cou^{22})-gellan fibers. For the PLO-gellan and PLO(Cou^{22})-fibers, the coumarin-containing sample exhibited higher breaking load (261 MPa) and strain (14%) values (Fig. 5B, curves 3) than the intact sample (139 MPa, 10%; Fig. 5B, curve 1). After a 30-min irradiation, the breaking stress of the irradiated sample was 340 MPa (Fig. 5B, curve 4) and this value was 1.3-fold higher than that of the non-irradiated sample (261 MPa, Fig. 5B, curve 3).

The above results from the uniaxially drawing test, together with those from the loop thread test, clearly indicate that the photochemical cross-linking between the side chain coumaryloxyacetyl groups in the polypeptides actually contribute to the reinforcement of the PIC fibers.

4. Conclusion

In conclusion, the present article described the synthesis of the coumarin-containing polypeptides using the water-soluble active ester method and the reinforcement of the PIC fiber utilizing the photochemical dimerization reaction between the pendent coumaryloxyacetyl groups. The toughness of the coumarin-containing PIC fibers is superior to those of the intact PLL- and PLO-gellan PIC fibers, and the mechanical strength of the coumarin-containing PIC fibers further increased upon irradiation for 30 min or less.

From these results, we can illustrate the reinforcement mechanism of the PIC fiber (Scheme 2). Fundamentally, the coumarin-containing PIC fibers are spun by a method similar to our earlier chitosan-gellan, PLL-gellan, chitosan-PLG, and PLL-PLG PIC fibers [4, 7]. The electrostatic interaction between the cationic polypeptide and the anionic gellan molecules forms the PIC fiber at the aqueous solution interface (Scheme 2A). The dropwise addition of the coumarin-containing polypeptide solution into the gellan solution forms the PIC fiber of which the inside is mostly comprised of the coumarin-containing polypeptide while the outside is gellan (Scheme 2B). The photochemical dimerization between the coumaryloxyacetyl groups forms the intra- and intermolecular covalent cross-linking. The cross-linking reaction significantly contributes to the mechanical reinforcement of the PIC fiber (Scheme 2C).



Scheme 2

In our ongoing study on PIC fibers composed of counter charged natural and related polymeric materials [4, 7], the present results proposed a new methodology for the reinforcement of PIC fibers. The characteristic features of the PIC fibers, which we have already reported, are as follows: i) a simple operation for spinning, ii) low-cost media (water, acetic acid, and alcohols), iii) enhanced tensile and knot strengths due to cross-linking agents, iv) dyeing with many colors [11, 13], v) environmental biodegradabilities [23], and vi) the adsorption of anionic medicines and endocrine disruptors [24]. The present results newly added vii) the photo-induced reinforcement of the mechanical strength by introducing the photoreactive group in the watery process. The combinations of these characteristics further inspire the multipurpose applications of PIC fibers.

Acknowledgements

This work was partly supported by Grants-in-aid for 21th Century COE Program and for Scientific Research (No. 12450330, No. 13555178, No. 13556057) by the Ministry of Education, Culture, Sports, Science and Technology of Japan.

References

1. K. OHKAWA, K. SHOUMURA, M. YAMADA, A. NISHIDA, H. SHIRAI and H. YAMAMOTO, *Macromol. Biosci.* **1** (2001) 149.
2. R. DIKIE, S. LABANA and R. BAUER (eds.), "Cross-Linked Polymers" (American Chemical Society, Washington, DC, 1988).
3. S. LABANA and R. DICKIE (eds.), "Characterization of Highly Cross-Linked Polymers" (American Chemical Society, Washington, DC, 1984).
4. H. YAMAMOTO and K. OHKAWA, in "Encyclopedia of Surface and Colloid Science," edited by A. Hubbard (Marcel Dekker, Inc, New York, 2002) p. 4242.
5. A. MUSTAFA, *Chem. Rev.* **51** (1952) 1.
6. H. YAMAMOTO, T. KITSUIKI, A. NISHIDA, K. ASADA and K. OHKAWA, *Macromolecules* **32** (1999) 1055.
7. K. OHKAWA and H. YAMAMOTO, in "Biopolymers," edited by A. Steinbuchel (Wiley-VCH, Weinheim, 2003) vol. 7, p. 465.
8. Y. KADOMA, A. UENO, K. TAKEDA, K. UNO and Y. IWAKURA, *J. Polym. Sci., Polym. Lett. Ed.* **12** (1974) 715.
9. Y. SUDA, Y. INAKI and K. TAKEMOTO, *J. Polym. Sci. Polym. Chem. Ed.* **21** (1983) 2813.
10. M. AMAIKE, Y. SENOO and H. YAMAMOTO, *Macromol. Rapid Commun.* **19** (1998) 287.
11. H. YAMAMOTO and Y. SENOO, *Macromol. Chem. Phys.* **201** (2000) 84.
12. K. OHKAWA, Y. TAKAHASHI, M. YAMADA and H. YAMAMOTO, *Macromol. Mater. Eng.* **286** (2001) 168.
13. H. YAMAMOTO, C. HORITA, Y. SENOO, A. NISHIDA and K. OHKAWA, *J. Appl. Polym. Sci.* **79** (2001) 437.
14. Y. TAKAHASHI, M. HACHISU, K. OHKAWA and H. YAMAMOTO, *Macromol. Rapid Commun.* **2002** (2002) 540.
15. K. OHKAWA, M. ANDO, Y. SHIRAKABE, Y. TAKAHASHI, M. YAMADA and H. YAMAMOTO, *Tex. Res. J.* **42** (2002) 120.
16. R. ANET, *Can. J. Chem.* **40** (1962) 1249.
17. G. S. HAMMOND, C. A. STOUT and A. A. LAMOLA, *J. Amer. Chem. Soc.* **86** (1964) 3103.
18. H. A. MORRISON, H. CURTIS and T. MCDOWELL, *ibid.* **88** (1966) 5415.
19. K. KOUGE, T. KOIZUMI, H. OKAI and T. KATO, *Bull. Chem. Soc. Jpn.* **60** (1987) 2409.
20. M. HATANO and M. YONEYAMA, *J. Amer. Chem. Soc.* **92** (1970) 1392.
21. R. E. FORNES, R. D. GILBERT, B. S. STOWE and G. P. CHEEK, *Tex. Res. J.* **43** (1973) 714.
22. G. A. HORSFALL, *ibid.* **52** (1982) 197.
23. K. OHKAWA, M. YAMADA, A. NISHIDA, N. NISHI and H. YAMAMOTO, *J. Polym. Environ.* **8** (2000) 59.
24. Y. TAKAHASHI, K. OHKAWA, M. ANDO, M. YAMADA and H. YAMAMOTO, *Macromol. Mater. Eng.* **286** (2001) 733.

Received 17 December 2002
and accepted 20 May 2003