

# Physical Characteristics of Silk Fibers Modified with Dibasic Acid Anhydrides

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## SYNOPSIS

The objective of this study was to investigate the physical properties of silk fibers modified with dibasic acid anhydrides. These are potentially attractive modifying agents to reduce the rate of photoyellowing of silk during and following UV irradiation. Several analytical techniques were employed, which included the measurement of the basic mechanical properties (tensile strength and elongation at break), equilibrium regain, amino acid analysis, dynamic viscoelastic measurements, X-ray diffractometry, and scanning electron microscopy (SEM). The succinylated silk fibers, which have been conditioned under different relative humidity atmospheres, always exhibited slightly higher equilibrium regain values than those of equivalently conditioned glutarylated silks. The amount of the basic amino acid residues slightly decreased following modification with both succinic and glutaric anhydrides. The birefringence values and the isotropic refractive indices decreased only slightly, which suggests that the fine structure of the treated silk fibers was not significantly altered. The X-ray diffraction curves demonstrated that no changes in the crystalline structure were induced by reaction with dibasic acid anhydrides. The tensile properties of the modified silks remained more or less unchanged. Only the initial tensile resistance of glutarylated silks in the dry state significantly decreased. The dynamic viscoelastic behavior of modified silk fibers was characterized by a reduced thermal stability. In fact, the onset temperature of the prominent  $E'$  peak corresponding to the molecular movement shifted to lower values. The surfaces of modified silk fibers were as smooth as that of the untreated control sample.

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## INTRODUCTION

In the broad use of silks for apparel textiles, problems are encountered associated with the yellowing of those fabrics with time when they are exposed to sunlight. Therefore, it is commercially relevant to provide an effective means of stabilizing silk fibers and fabrics to photodegradation and, concurrently, to also improve the easy-care characteristics of such silk fabrics. Previous efforts to achieve these objectives often resulted in the degradation of the silk

with the loss of the original fiber's desirable features. This consequence detracts from their potential viability.

In our laboratories, treatments of *Bombyx mori* silks involving the chemical modification<sup>1-3</sup> and polymerization of particular monomers<sup>4-9</sup> within the fiber have shown considerable promise in improving some minor textile performances, reducing the rate of photoyellowing, and loss of whiteness while sustaining the often-lost desirable mechanical properties and handle.

Among many chemical agents for reducing the rate of photoyellowing, epoxides<sup>1,3</sup> and dibasic acid anhydrides,<sup>10-12</sup> which act to produce intermolecular cross-links, as well as selected vinyl monomers,<sup>13</sup>

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appeared potentially promising important agents for these objective purposes. The effectiveness of the epoxide treatment in retarding the photoyellowing of silk is still inadequate for its satisfactory commercial implementation.

In a recent paper,<sup>13</sup> we studied the polymerization of *N*(*n*-butoxymethyl) methacrylamide (BMA) within silk and the characteristics of the product were examined. Among the product properties examined, we observed a marked effect of poly(BMA) loaded onto silk in maintaining the whiteness and reducing the rate of yellowing following UV irradiation.

Some of the authors extensively studied the reaction of silk with dibasic acid anhydrides.<sup>10-12</sup> These modifying agents were highly effective in producing substantial modifications of the dyeing properties of silk fibers without affecting their textile performances.<sup>10,12</sup> Furthermore, glutarylation has been found to protect silk fibers from yellowing following UV irradiation.<sup>10</sup> We considered, therefore, that dibasic acid anhydrides have potentially promising applications as an effective means for materially improving silk fibers.

In this study, we report the equilibrium regain, the amino acid analysis, the refractive indices, the mechanical properties, the viscoelastic behavior, and the crystalline structure of silk fibers modified with dibasic acid anhydrides, i.e., succinic and glutaric anhydrides. With significant basic experience in polymer science and textile science (particularly on protein fibers), as well as physical techniques of polymer analysis, positive contributions could be made to characterize the chemically modified silk fibers in view of their industrial exploitation.

## EXPERIMENTAL

### Materials

Reagent-grade succinic and glutaric anhydrides, purchased from Wako Pure Chemical Industries Ltd., were used without further purification. Degummed and bleached silk fibroin fibers (21d/1 × 2) were treated with 10% (w/v) succinic and glutaric anhydrides in dimethylformamide (DMF) for different times. The reaction vessel was attached to a reflux condenser and held in a thermostated bath at 75°C. The material-to-liquor ratio of 1 : 20 was maintained. At the end of the required treatment times, the samples were washed with isopropanol, then with acetone at 55°C for 1 h to remove the untreated anhydride and successively with tap water.

The treated samples were finally dried at 110°C and weighed.

### Measurements

The moisture regain was determined on dried samples kept for 7 days at 20°C in environments with different but fixed relative humidities (65, 84, and 88% RH). Regain values were expressed as grams of moisture/10 g of fibroin.

Silk fibroin fibers modified with dibasic acid anhydrides were hydrolyzed by heating at 110°C for 20 h in HCl 6*N* under vacuum. The hydrolysates were filtered through a glass disk (No. 6) to remove small residues, dried in rotary evaporator at 40°C, and dissolved in a buffer solution at pH 2.2 (1 mg/L). The amino acid composition was determined using a Hitachi L-8500 rapid amino acid analyzer.

The refractive indices of modified silk fibers were measured by the Becke's line method using a polarization microscope under monochromatic light (Na light) at 20°C and 65% RH as previously described.<sup>8</sup> X-ray diffraction measurements were performed with a Rigaku Denki Co. RU-200 diffractometer, using CuK $\alpha$  radiation.

Fiber strength and elongation at break were measured on single threads with an automatic Tensilon tensile tester (UTM-II) in standard conditions at 20°C and 65% RH and in water. The rate of strain was 20 mm/min on fixed sample lengths of 50 mm. Each recorded value was the average of 20 results.

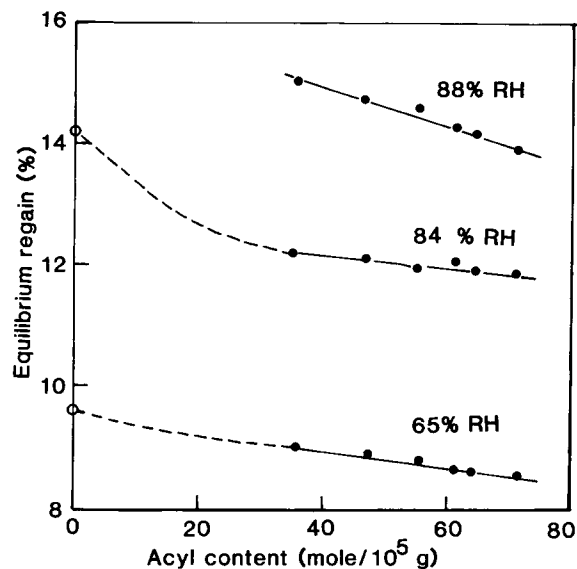
The dynamic storage ( $E'$ ) and loss ( $E''$ ) moduli of silk yarns were measured at 10 Hz using a Toyoseiki Rheograph Solid-S. The temperature range was from room temperature up to 250°C with the sample heated at 2°C/min. The sample length was 15 mm under an applied initial tensile load of 30 g.

The surface morphology of the modified silk fibers was observed with a Jeol JAX-333S scanning electron microscope after gold coating.

## RESULTS AND DISCUSSION

### Equilibrium Regain

Equilibrium regains of silk fibers modified with dibasic acid anhydrides were measured under different relative humidities and plotted as a function of the acyl content (Fig. 1). The equilibrium regains of untreated control samples increased with the relative humidity of the atmosphere. The values of untreated control silk fibers were 9.7 and 14.2% at 65 and 84% RH, respectively. As the acyl content increased, the



**Figure 1** Equilibrium regain of silk fibers modified with dibasic acid anhydrides measured under different relative humidity conditions (65, 84, and 88% RH) as a function of the acyl content. (●) Untreated control silk fibers.

equilibrium regain of both succinylated and glutarylated silk fibers linearly decreased, as shown by the behavior of the curves in Figure 1. We observed that silk fibers modified with succinic anhydride always exhibited an equilibrium regain slightly higher than that of glutarylated silk fibers, suggesting that the latter achieved a bit higher hydrophobic character after chemical modification. These data are consistent with the existence of additional free acyl groups in silk fibers treated with succinic anhydride. The lower content of free acyl groups of glutarylated silk should be ascribed mainly to the involvement of this modifying agent in the formation of intermolecular cross-links.<sup>10</sup>

### Amino Acid Analysis

The reactivity toward silk fibroin of succinic and glutaric anhydrides was evaluated by determination of the amino acid composition of modified silk fibers (Table I). In previous papers<sup>10-12</sup> we reported that the main reactive sites for acylation of silk fibroin are the free amino groups of the basic amino acid residues (lysine, histidine, and arginine). Their amount was found to decrease as the acyl content increased.<sup>11</sup> The data reported in Table I confirmed this behavior and showed that the concentration of basic amino acid residues is lower in both glutarylated and succinylated silk fibers, especially at high weight gains (samples G-3 and S-3). Hydroxyl

groups of serine, threonine, and tyrosine should be included among the reactive sites for silk acylation. However, their content did not change significantly because the ester bond once formed between silk fibroin and the dibasic acid anhydrides was easily broken during acid hydrolysis.

### Refractive Indices

Table II lists the birefringence ( $\Delta n$ ) and isotropic refractive index ( $n_{\text{ISO}}$ ) values of silk fibers modified with glutaric and succinic anhydrides as a function of the acyl content. The birefringence of both glutarylated and succinylated silk fibers remained almost unchanged regardless of the chemical modification. The isotropic refractive index values, however, showed a slight decrease following the acylation reaction and then attained a constant value as the weight gain increased above 6–7%. Since  $\Delta n$  and  $n_{\text{ISO}}$  may be regarded as a measure of the average molecular orientation and crystallinity of silk fibers, respectively, we can conclude that the chemical modification with dibasic acid anhydrides did not significantly affect the physical structure of the fibers. Similar results were obtained using itaconic anhydride as the modifying agent.<sup>11</sup>  $\Delta n$  and  $n_{\text{ISO}}$  values showed a different trend in relation to the increase of weight gain, but the extent of change was very small. These findings suggest that the chemical

**Table I** Amino Acid Composition of Untreated Control, Succinylated (S), and Glutarylated (G) Silk Fibers

Amino Acids (mol %)	Control	S-1	S-3	G-1	G-3
Asp	1.65	1.60	1.62	1.60	1.62
Thr	0.85	0.91	0.89	0.84	0.89
Ser	10.62	10.79	10.68	10.54	10.54
Glu	1.21	1.14	1.24	1.19	1.22
Gly	45.09	44.55	45.09	45.22	45.20
Ala	29.21	29.23	29.18	29.22	29.18
Val	2.49	2.40	2.43	2.53	2.52
Met	0.14	0.18	0.14	0.19	0.18
Ile	0.71	0.69	0.68	0.68	0.71
Leu	0.54	0.58	0.53	0.54	0.55
Tyr	4.97	5.17	5.03	5.01	5.00
Phe	0.70	0.86	0.71	0.70	0.72
Lys	0.33	0.32	0.27	0.31	0.28
His	0.18	0.19	0.19	0.17	0.17
Arg	0.49	0.46	0.40	0.48	0.40
Pro	0.77	0.97	0.88	0.74	0.79

Weight gains: S-1 6.4%; S-3 11.6%; G-1 7.6%; G-3 13.3%.

**Table II Physical Properties of Silk Fibers Modified with Succinic and Glutaric Anhydrides**

Sample	Treatment Time (min)	Weight Gain (%)	Acyl Content (mol/10 <sup>5</sup> g)	$\Delta n$	$n_{ISO}$
Control	0	0	0	0.049	1.554
Succinylated					
S-1	45	6.4	55	0.046	1.553
S-2	90	8.8	75	0.046	1.553
S-3	270	11.6	99	0.046	1.552
Glutarylated					
G-1	45	7.6	58	0.0455	1.552
G-2	90	10.4	79	0.046	1.5523
G-3	270	13.3	102	0.045	1.551

$\Delta n$ : birefringence;  $n_{ISO}$ : isotropic refractive index.

modification by acylation with dibasic acid anhydrides can be considered a mild treatment from the physical point of view, since it does not induce significant changes in the fine structure of silk fibers.

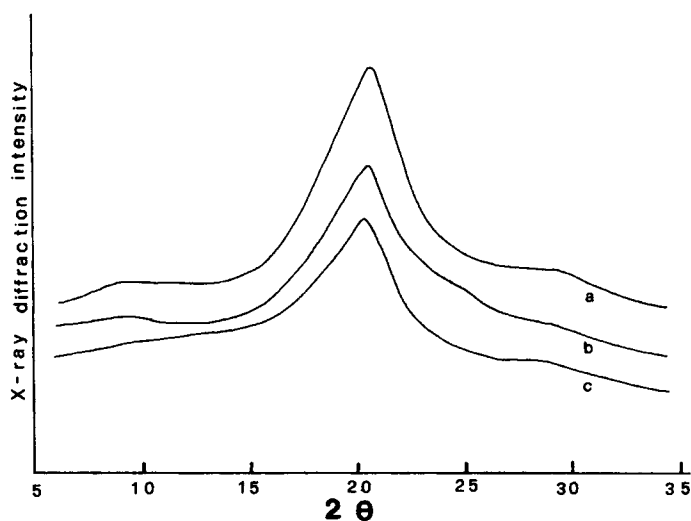
### X-ray Diffraction Curves

To ascertain if some changes in the crystalline structure were induced by the chemical modification reaction, X-ray diffraction curves of modified silk fibers were measured (Fig. 2). The untreated control sample (a) exhibited a major X-ray diffraction peak at 20.5° corresponding to the specific crystalline spacing of 4.39 Å for silk fibers with high molecular orientation. The succinylated (b) and glutarylated (c) silk fibers with a weight gain of 11.6 and 13.3%,

respectively, showed similar X-ray diffraction patterns. The above results demonstrated that the crystalline structure of silk fibers with oriented  $\beta$  crystals was not affected by the modification reaction and that dibasic acid anhydrides bound to the reactive sites in the amorphous regions.

### Mechanical Properties

The mechanical properties of silk fibers modified with dibasic acid anhydrides were measured either in the dry state (standard conditions: 20°C, 65% RH) or in water. The results are listed in Table III. As expected, the tensile strength was lower in the wet than in the dry state (-29%). The values obtained on acylated silk fibers remained almost un-



**Figure 2** X-ray diffraction intensity curves of (a) untreated control, (b) succinylated, and (c) glutarylated silk fibers. Weight gain: (b) 11.6%; (c) 13.3%.

**Table III Mechanical Properties in the Dry State (Standard Conditions: 20°C, 65% RH) and in Water of Untreated Control, Succinylated (S), and Glutarylated (G) Silk Fibers**

Sample	Strength (g/d) <sup>a</sup>		Elongation (%)		Tensile Resistance <sup>b</sup> (g/d/% × 100)	
	Dry	Wet	Dry	Wet	Dry	Wet
Control	3.5	2.5	12.3	23.5	77.9	21.2
Succinylated						
S-1	3.8	2.6	14.3	20.8	79.2	20.0
S-2	3.6	2.6	13.5	21.1	81.2	19.7
S-3	3.4	2.5	14.5	22.3	79.4	18.1
Glutarylated						
G-1	3.6	2.7	14.0	19.3	71.6	21.3
G-2	3.5	2.6	13.4	19.1	69.3	20.2
G-3	3.4	2.5	12.9	19.0	73.5	19.1

Weight gains: S-1 6.4%; S-2 8.8%; S-3 11.6%; G-1 7.6%; G-2 10.4%; G-3 13.3%.

<sup>a</sup> 1 g/d =  $1.22 \times 10^3$  kgf/cm<sup>2</sup>.

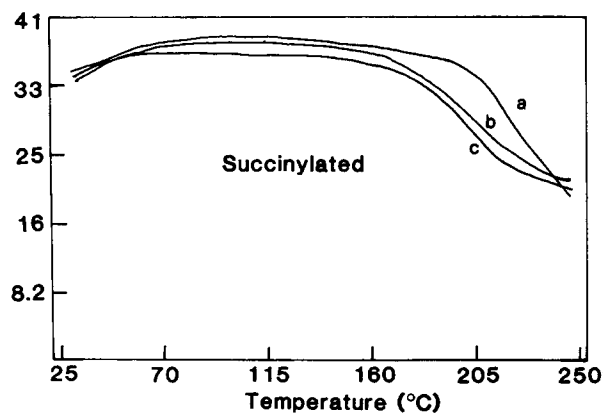
<sup>b</sup> The initial tensile resistance is a measure corresponding to the Young's modulus.

changed compared with the untreated control sample. The slight changes registered were independent of the modifying agent used and of the weight gain attained. This observation is consistent with the fact that no significant changes of the fine structure were detected by the refractive index and X-ray diffraction measurements (Table II and Fig. 2). The elongation at break of modified silk fibers slightly increased in the dry state, whereas it showed a small but constant decrease in the wet state, which was more evident for glutarylated silk fibers (−19%). The initial tensile resistance, evaluated from the slope of the stress–strain curves, was significantly lower in the wet than in the dry state (−73%) for both control and modified silk fibers. Among the latter, it is worth noting the behavior of the glutarylated samples, whose initial tensile-resistance values measured in the dry state decreased following the chemical modification. These results of the mechanical properties seem to support the hypothesis of intermolecular cross-link formation by reaction of glutaric anhydride within silk fibroin fibers.<sup>10</sup>

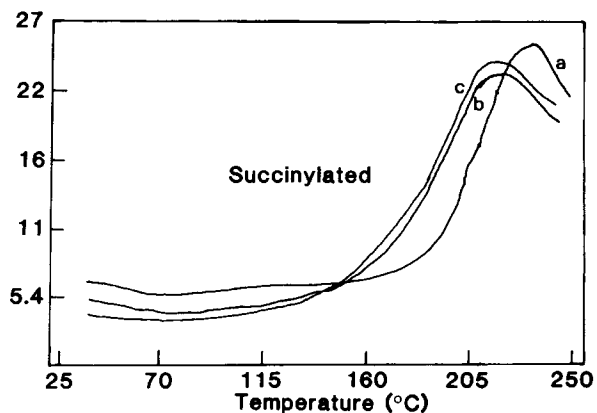
### Dynamic Viscoelastic Behavior

Figures 3 and 4 show the behavior of the dynamic storage ( $E'$ ) and loss ( $E''$ ) moduli of succinylated silk fibers with different weight gains, respectively. The onset temperature at which the dynamic storage modulus of the untreated control sample began to decrease is located at around 200°C [Fig. 3(a)]. The corresponding values for modified silk fibers (b,c)

shifted to lower temperature, giving evidence of their reduced thermal stability. This behavior is noticeable even at relatively low weight gain (b), the thermal movement of fibroin molecules becoming evident at about 170–180°C. The loss modulus peak of untreated control silk fibers [Fig. 4(a)] showed a maximum at 230°C, which shifted down to 215 and 208°C for succinylated fibers with 6.4 and 11.6% weight gain, respectively (b,c). Furthermore, the  $E''$  peaks of modified fibers became broader, their onset temperature being located at around 145°C, about 35°C lower than that of the untreated control sample.



**Figure 3** Dynamic storage modulus ( $E'$ ) curves of (a) untreated control and (b,c) succinylated silk fibers. Weight gain: (b) 6.4%; (c) 11.6%.

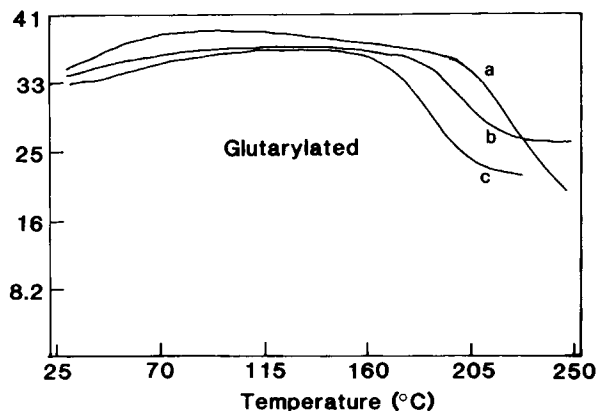


**Figure 4** Dynamic loss modulus ( $E''$ ) curves of (a) untreated control and (b,c) succinylated silk fibers. Weight gain: (b) 6.4%; (c) 11.6%.

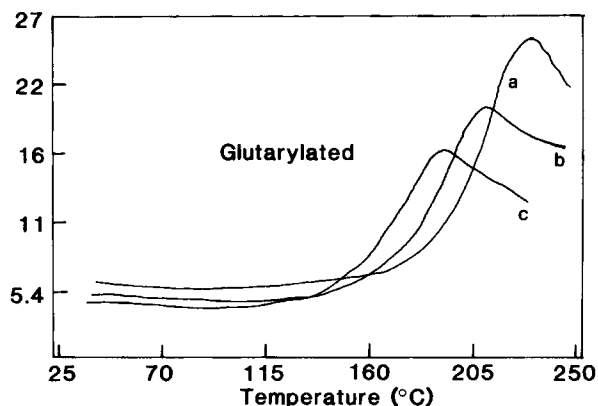
The observed  $E'$  and  $E''$  curves of glutarilated silk fibers are shown in Figures 5 and 6, respectively. Their viscoelastic behaviors exhibited a striking resemblance to those of succinylated fibers.

It is reasonable to assume that dibasic acid anhydrides react toward the active sites of the amorphous regions as well as toward the laterally ordered regions located in the fringed fibril volumes at the end of each individual ordered element aligned along the fiber axis. The crystalline regions cannot be penetrated by the modifying agents, as demonstrated by the X-ray diffraction patterns (Fig. 2).

It has been reported that the prominent  $E''$  peak at around  $230^{\circ}\text{C}$  of *Bombyx mori* silk fibroin can be attributed to the thermal movements in the ordered volumes of the crystalline regions, because the spacing [ $d_{(200)}$ ] corresponding to the intersheet distance



**Figure 5** Dynamic storage modulus ( $E'$ ) curves of (a) untreated control and (b,c) glutarilated silk fibers. Weight gain: (b) 7.6%; (c) 13.3%.



**Figure 6** Dynamic loss modulus ( $E''$ ) curves of (a) untreated control and (b,c) glutarilated silk fibers. Weight gain: (b) 7.6%; (c) 13.3%.

of the  $\beta$ -structure is observed to gradually expand at above  $180^{\circ}\text{C}$ ,<sup>14</sup> at which temperature the  $E''$  value began to increase (Fig. 5).

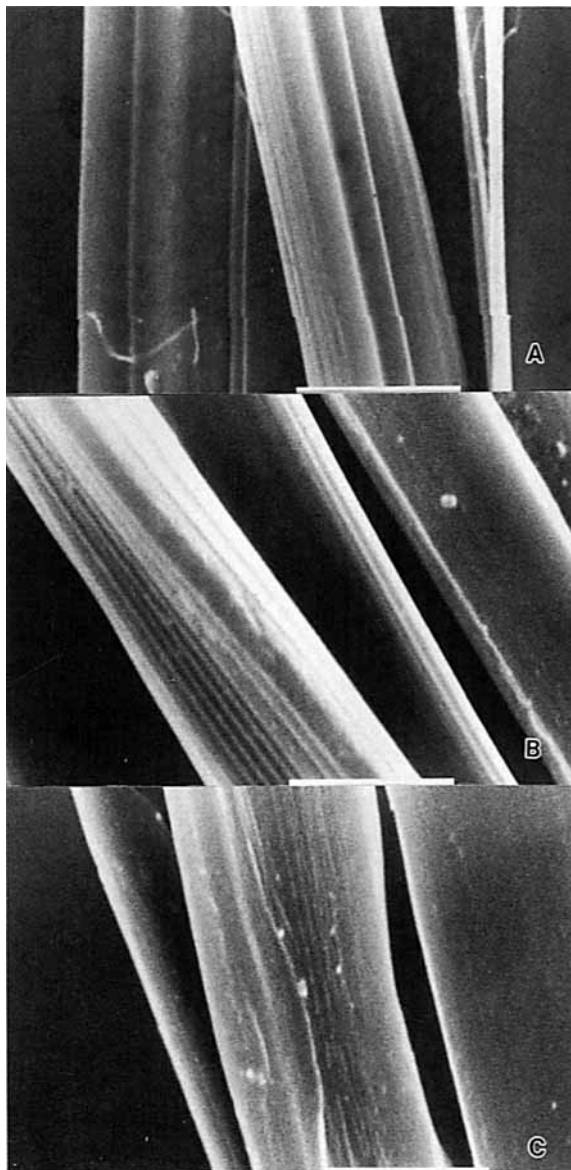
The described viscoelastic behavior of silk fibers modified with dibasic acid anhydrides implies that the thermally induced molecular movements in the overall ordered regions of silk fibroin fibers are facilitated by the chemical modification reaction. It is therefore concluded that the thermal movement of the ordered regions should have been greatly influenced by the changes induced in the amorphous regions, within which the modifying agent molecules bound to silk fibroin chains. In fact, as shown by the X-ray diffraction data (Fig. 2), the crystalline regions were not directly affected by the acylation reaction.

### Surface Characteristics

The surfaces of silk fibers modified with dibasic acid anhydrides were examined by scanning electron microscopy (SEM). SEM micrographs of both succinylated [Fig. 7(b) and (c)] and glutarilated [Fig. 8(b) and (c)] fibers showed that their surfaces were as smooth as those of the untreated control samples (a). It is therefore concluded that the reaction with the modifying agents did not affect the surface characteristics of silk fibers, keeping their original lustrous appearance unchanged.

### CONCLUSIONS

Results reported in this paper show that the physical properties of silk fibers modified with dibasic acid

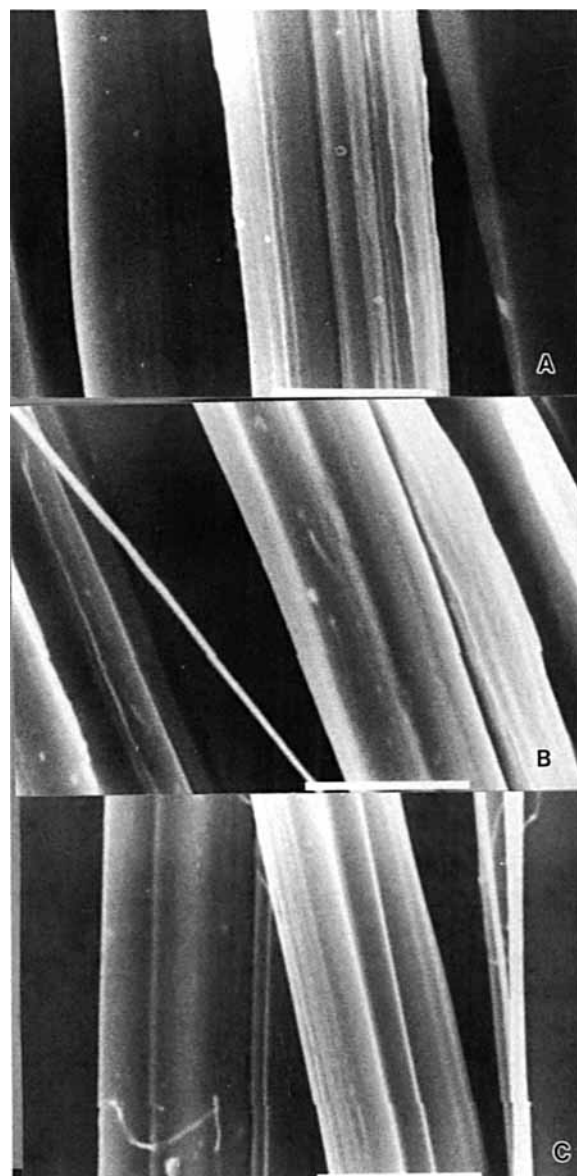


**Figure 7** SEM micrographs of (A) untreated control and (B,C) succinylated silk fibers. Weight gain: (B) 6.4%; (C) 11.6%.

anhydrides are largely unaffected by the chemical reaction. Fine structural features, such as molecular orientation and crystallinity, remained unchanged, while thermal stability was only slightly reduced. Mechanical properties and surface characteristics, which determine textile performances and appearance (luster) of silk products, are very similar to untreated silk fibers.

In previous papers,<sup>10-12</sup> we demonstrated the effectiveness of dibasic acid anhydrides as modifying

agents for reducing the rate of photodegradation and improving dye affinity of modified silk fibers. It was also demonstrated<sup>10</sup> that modification with dibasic acid anhydrides is characterized by treatment conditions milder than those used for other chemical modification techniques, such as grafting with vinyl monomers. On the basis of the present results, we can envisage the possibility of exploiting this modification technique for the development of new silk goods, either pure or blended with synthetic fibers,



**Figure 8** SEM micrographs of (A) untreated control and (B,C) glutarylated silk fibers. Weight gain: (B) 7.6%; (C) 13.3%.

and renew traditional dyeing and printing techniques.

## REFERENCES

1. M. Tsukada, H. Shiozaki, H. Urashima, and M. Yokozawa, *J. Text. Inst.*, **80**, 547 (1989).
2. M. Tsukada, H. Shiozaki, and A. Konda, *J. Appl. Polym. Sci.*, **41**, 1213 (1990).
3. M. Tsukada, M. Nagura, H. Ishikawa, and H. Shiozaki, *J. Appl. Polym. Sci.*, **43**, 643 (1991).
4. M. Tsukada, *J. Seric. Sci. Jpn.*, **53**, 380 (1984).
5. M. Tsukada, *J. Appl. Polym. Sci.*, **35**, 2133 (1988).
6. M. Tsukada, *J. Appl. Polym. Sci.*, **35**, 965 (1988).
7. M. Tsukada and H. Shiozaki, *J. Appl. Polym. Sci.*, **39**, 1289 (1990).
8. M. Tsukada, T. Yamamoto, N. Nakabayashi, H. Ishikawa, and G. Freddi, *J. Appl. Polym. Sci.*, **43**, 2115 (1991).
9. M. Tsukada, G. Freddi, M. Matsumura, H. Shiozaki, and N. Kasai, *J. Appl. Polym. Sci.*, **44**, 799 (1992).
10. M. Tsukada and H. Shiozaki, *J. Appl. Polym. Sci.*, **37**, 2637 (1989).
11. M. Tsukada, Y. Gotoh, G. Freddi, H. Shiozaki, and H. Ishikawa, *J. Appl. Polym. Sci.*, **45**, 1719 (1992).
12. M. Tsukada, Y. Gotoh, G. Freddi, and H. Shiozaki, *J. Appl. Polym. Sci.*, **45**, 1189 (1992).
13. M. Tsukada, H. Shiozaki, and J. S. Crighton, *J. Appl. Polym. Sci.*, to appear.
14. M. Nagura, K. Gotoh, and H. Ishikawa, *Kobunshi Ronbunshu*, **34**, 389 (1977).

Received September 16, 1992

Accepted June 18, 1993