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X-ray and Electron Diffraction Study of Cellulose Crystal Structures[*1]

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X선 및 전자선회절법에 의한 천연셀룰로오스의 결정구조 해석[*1]

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Cellulose I 에서 Cellulose II 로의 결정변태기구를 X선 및 전자선 회절법과 현미경적 방법을 이용하여 구명하였다.

X선 회절 결과, Na-cellulose I을 고온에서 수세할 경우 Cellulose I 과 Cellulose II 의 혼합형 회절도가, 저온에서 수세할 경우 Na-cellulose IV의 회절도가 얻어졌다.

전자선회절 결과, 고온수세의 시료는 Cellulose I 과 Cellulose II 의 혼합형이, 저온수세의 시료는 Cellulose II 의 회절도가 얻어졌다. 또한 고온수세 시료의 전자선회절도로부터 섬유벽의 내측부가 외측부보다 재생 Cellulose I 의 양이 많은 것이 확인되었다.

따라서 알칼리 팽윤시 섬유벽내에는 불완전한 팽윤이 발생하는데 그 정도는 내측부가 더욱 심한 것으로 생각된다. 이때 형성되는 불완전한 Na-cellulose I 은 고온수세의 경우는 탈수에 의해 Cellulose I로, 저온수세의 경우는 수화에 의해 Cellulose II로 변태되지만 완전히 팽윤된 Na-cellulose I 은 Cellulose I 로 재생될 수 없는 것으로 생각된다. 현미경적 실험결과, mercerization과정에서 cellulose 분자쇄의 packing이나 conformation의 변화와 관련하여 microfibril의 흐트러짐은 발생하지 않는 것으로 생각되었다.

Keywords : Cellulose I, cellulose II, mercerization, Na-cellulose I, ramie

1. INTRODUCTION

The crystal transformation of cellulose I to cellulose II passes through Na-cellulose I. On the transformation mechanism of cellulose I to cellulose II, Okano and Sarko(1985), and Nishimura and Sarko(1987) reported that the transformation of cellulose I to cellulose II occurred at the stage of Na-cellulose I formation by the intermingling of "up" and "down" cellulose chains. On the other hand, Hayashi *et al.*(1974) suggested that the transformation occurred in the stage of washing. They reported that Na-cellulose I can be classified into Na-cellulose I_I(reversible) and Na-cellulose I_{II} (irreversible). The reversibility of Na-cellulose I to cellulose I is an important problem to understand the mechanism of mercerization.

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On the regeneration of Na-cellulose I to cellulose I, attentions have been paid to the influence of washing temperature. Sisson and Saner(1941) reported that Na-cellulose I from various cotton origins was partially regenerated to cellulose I by high temperature washing, and suggested that both the native cellulose and the mercerized cellulose exist in partially mercerized fibers as two separate phases, which are localized in the fiber. Recently, Hayashi *et al.*(1976) examined the relationship between the washing temperature and the reconversion of Na-cellulose I into cellulose I, and found that the percentage of reconversion into cellulose I increased with increasing washing temperature. The results were explained in terms of "memorial phenomenon" (Hayashi's terminology), involving that a part of Na-cellulose I had a "bent" type chain conformation which can be reconverted into cellulose I. In previous paper, however, Kim *et al.*(1990) reported that Na-cellulose I_I was reconverted into cellulose I through washing with water and the regenerated cellulose I was localized heterogeneously within a fiber wall caused by a gradient of swelling.

Consequently, in the present study, attention was directed towards tracing the influence of washing temperature on the reversibility of Na-cellulose I by X-ray and electron diffraction analysis methods. Polarized and electron microscopy were also used to investigate the difference of orientation of microfibrils in native and alkali-treated ramie fibers.

2. MATERIALS & METHODS

2.1 Materials

Bundles of 100 purified ramie fibers (cellulose I, *Boehmaria nivea* Gaud.) and mercerized ramie fibers(cellulose II) were used as starting materials. They were fastened in a Teflon specimen holder to ensure a strict alignment along the fiber axis. Mercerized ramie fibers were prepared by immersing them in 3.5N sodium hydroxide solution, and the treatment was repeated until the X-ray diffraction diagrams of them showed complete conversion to cellulose II.

2.2 Mercerization

Mercerization was conducted for 2 hours at room temperature using 3.5N sodium hydroxide solution. These Na-cellulose I samples were used to take the X-ray diagrams, and then washed for 1 hour with distilled water at 0℃ and 100℃, respectively.

2.3 X-ray diffraction analysis

X-ray diffraction diagrams of Na-cellulose I and regenerated cellulose samples were recorded using a vacuum camera mounted on a Rigaku X-ray generator(RU-200BH). X-ray diffractograms were also recorded by symmetrical transmission mode using JEOL-JDX5B diffractometer. Both X-ray diffraction diagrams and diffractograms of wet samples were obtained by wrapping them in Mylar to prevent solvent evaporation during recording.

2.4 Polarized and electron microscopy

The samples subjected to X-ray diffraction analysis were embedded in epoxy resin(Spurr, 1969). Longitudinal ultrathin sections(ca. 100nm thickness) and semithin sections(ca. 1,000nm thickness) were cut with a LKB ultramicrotome equipped with a diamond knife. Semithin sections were examined under a polarized microscope, and ultrathin sections were mounted on Cu grids with Formval supporting films. After removal of epoxy resin of ultrathin sections according to the modified Mayor's method(Fujii *et al.*, 1987), the sections were stained negatively by 1% aqueous uranyl acetate. The selected area diffraction diagrams were taken from 2㎛ regions of internal and exteral parts of a fiber wall. Observation and electron diffraction were performed on JEM-100CX and JEM-2000EXII electron microscopes operating at 100kV and 200kV, respectively.

3. RESULTS & DISCUSSION

3.1 Reconversion from Na-cellulose I to cellulose I in relation to washing temperature

When the ramie cellulose I was mercerized with 3.5N sodium hydroxide solution for 2 hours, the X-ray diffraction diagram did not show any reflections derived from cellulose I, but showed only Na-cellulose II(Fig. 1A). Similarly cellulose I_I was integrally converted to Na-cellulose I_{II}(Fig. 1B) by the same treatment.

The X-ray diffraction diagrams in Fig. 2 were recorded from Na-cellulose I_I which was washed at 0℃ (A) and 100℃ (B). The diagrams were characterized clearly as Na-cellulose IV(A) and as a mixture of Cellulose I and Cellulose II(B).

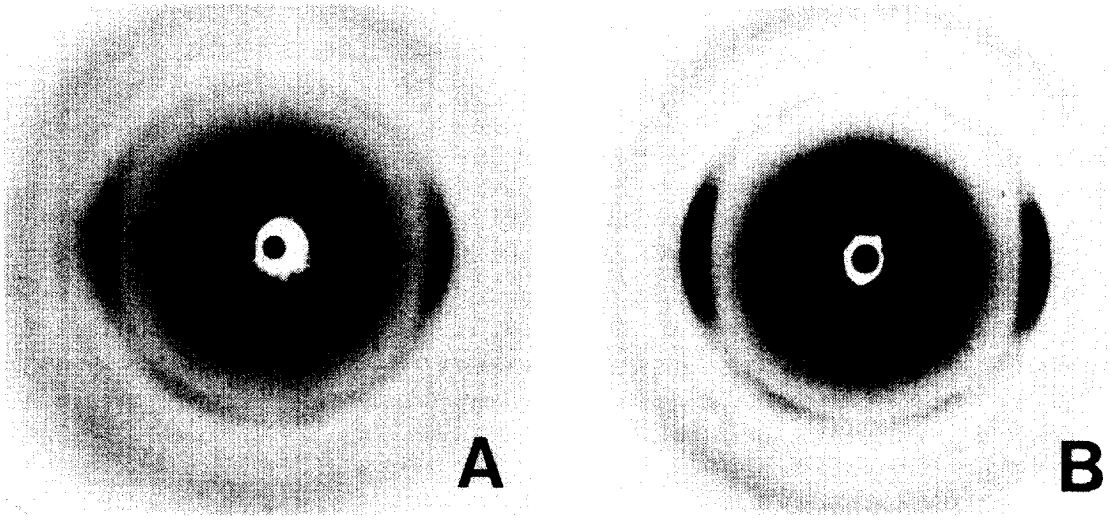

Fig. 1. X-ray diffraction diagrams of Na-cellulose I_I(A) and Na-cellulose I_{II}(B). Fiber axis is vertical.

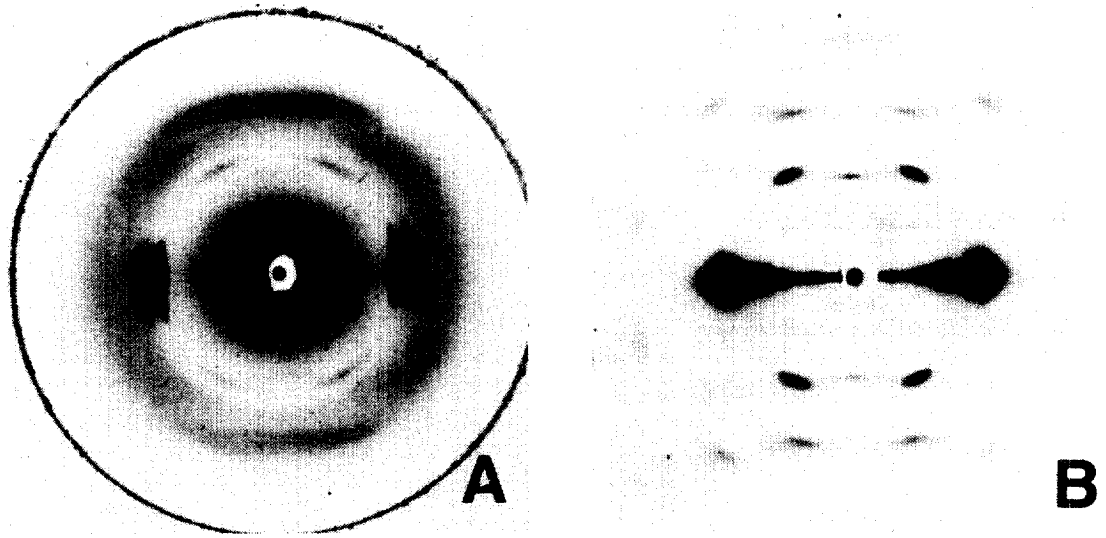

Fig. 2. X-ray diffraction diagrams obtained after washing Na-cellulose I_I with water at 0℃(A) and 100℃(B). Fiber axis is vertical.

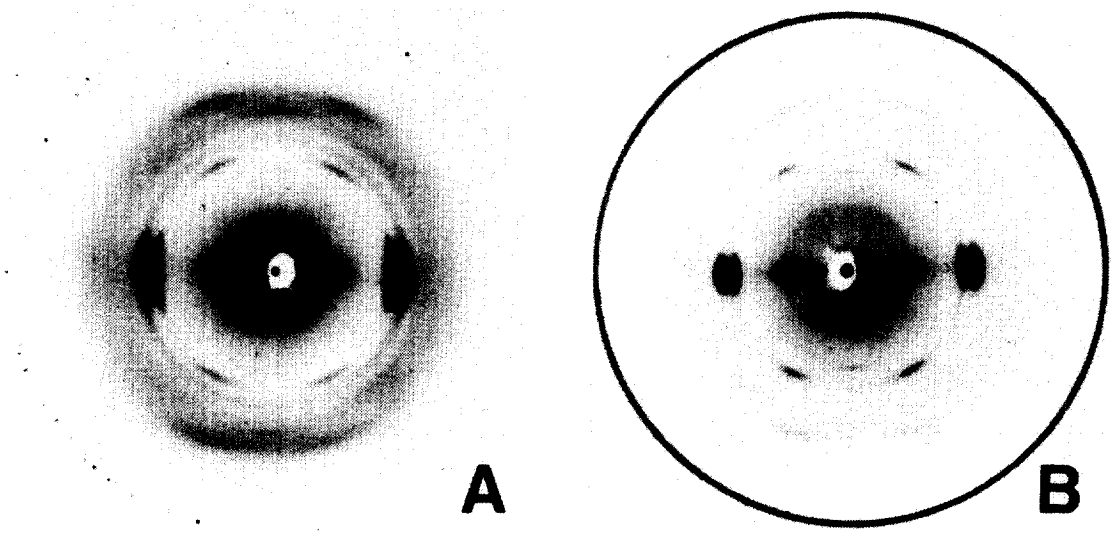

Fig. 3 X-ray diffraction diagrams obtained after washing Na-cellulose I_{II} with water at 0℃(A) and 100℃(B).

respectively. Fig.2 indicated that washing of Na-cellulose I_I at 0℃ followed by drying resulted in a complete formation of cellulose II and disappearance of Cellulose I. In addition, the Na-cellulose IV obtained after washing at 0℃ could not be reconverted into cellulose I even after long time washing at 100℃. Fig. 3 was recorded from Na-cellulose I_{II} which was washed at 0℃(A) and 100℃(B). The diagrams were characterized clearly as Na-cellulose IV(A) and as cellulose II(B). These results indicated that Na-cellulose IV once completely formed cannot be reconverted into cellulose I.

Hayashi *et al.*(1976) reported that regenerated cellulose I increased with increasing washing temperature. They interpreted this phenome-

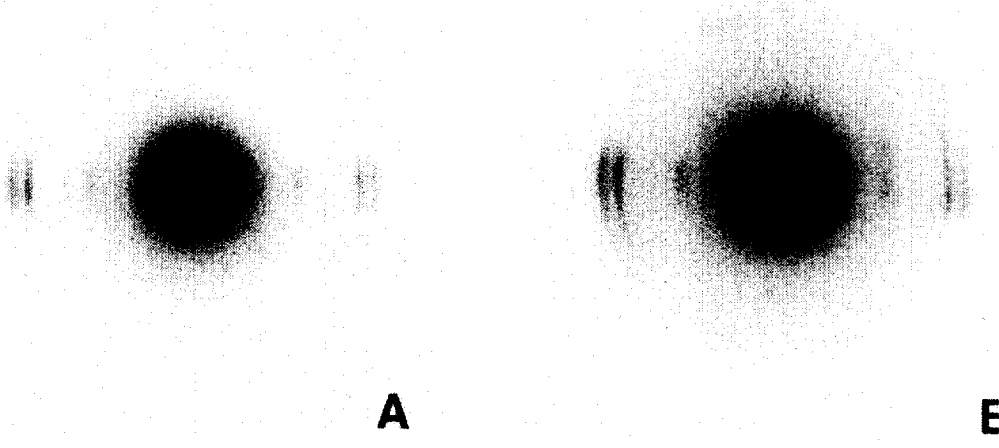

Fig. 4 Electron diffraction diagrams of external(A) and internal(B) layers after washing Na-cellulose I_I with water at 0℃.

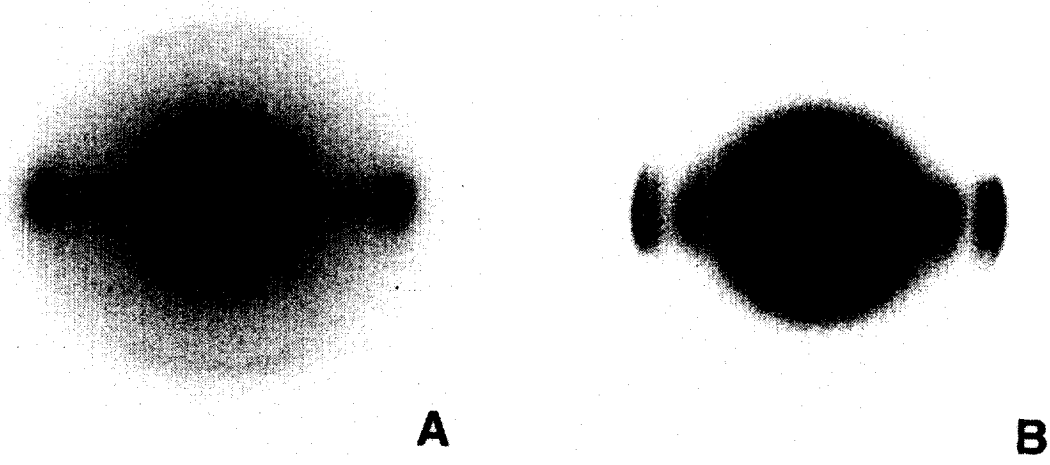

Fig. 5. Electron diffraction diagrams of external(A) and internal(B) layers after washing Na-cellulose I_I with water at 100℃.

non as the conversion of chain conformation from "bent form Na-cellulose I_I(reversible)" into "bent-twisted Na-cellulose I_{II} (irreversible)".

The present work clearly showed that when the Na-cellulose I_I and I_{II} were washed at 100 ℃, they were transformed directly into a mixture of cellulose I and cellulose II, and cellulose II, respectively, without the formatiom of Na-cellulose IV.

3.2 Electron diffraction analysis of ramie fibers

Electron diffraction diagrams(Figs. 4, and 5) were obtained from the external(a) and internal(b) layers of ramie fibers at 0℃ and 100℃, respectively. The diagrams of Fig. 5 show the

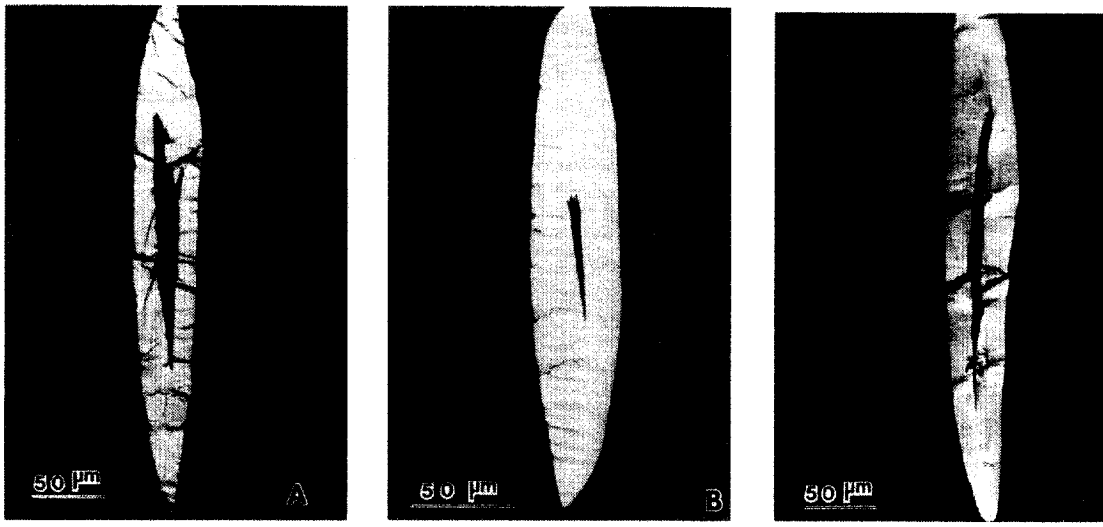

Fig. 6. Polarized micrographs of cellulose I(A), a mixture of celluose I and cellulose II (B) and cellulose II (C).

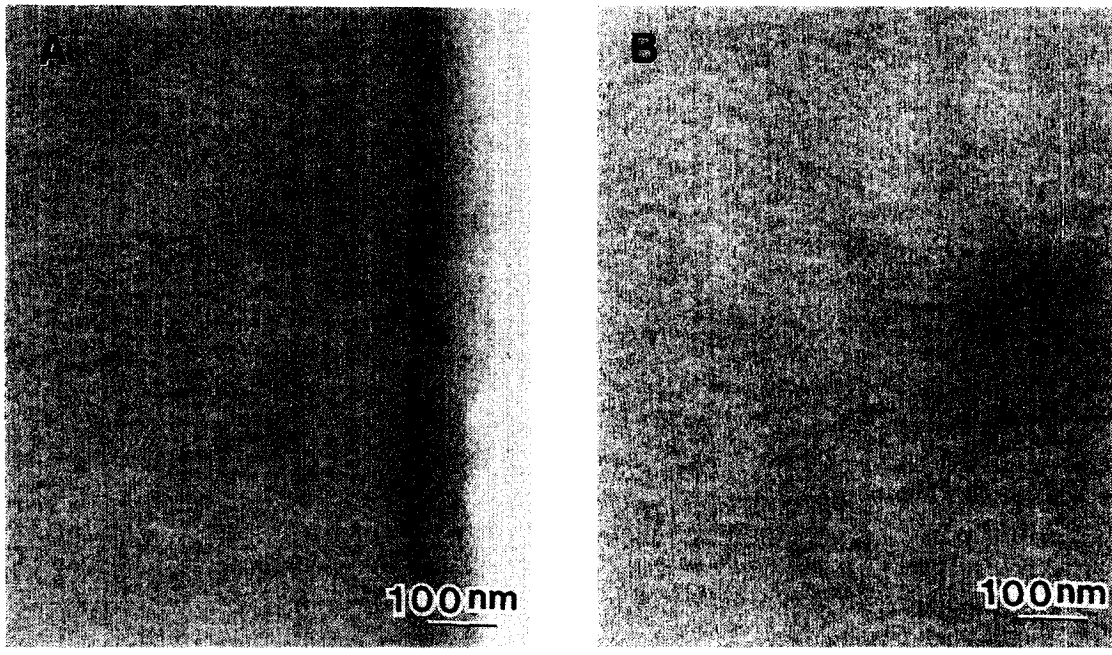

Fig. 7. Electron micrographs of cellulose I.
Notes: (A) : Outer part of a fiber wall. (B) : Inner part of a fiber wall.

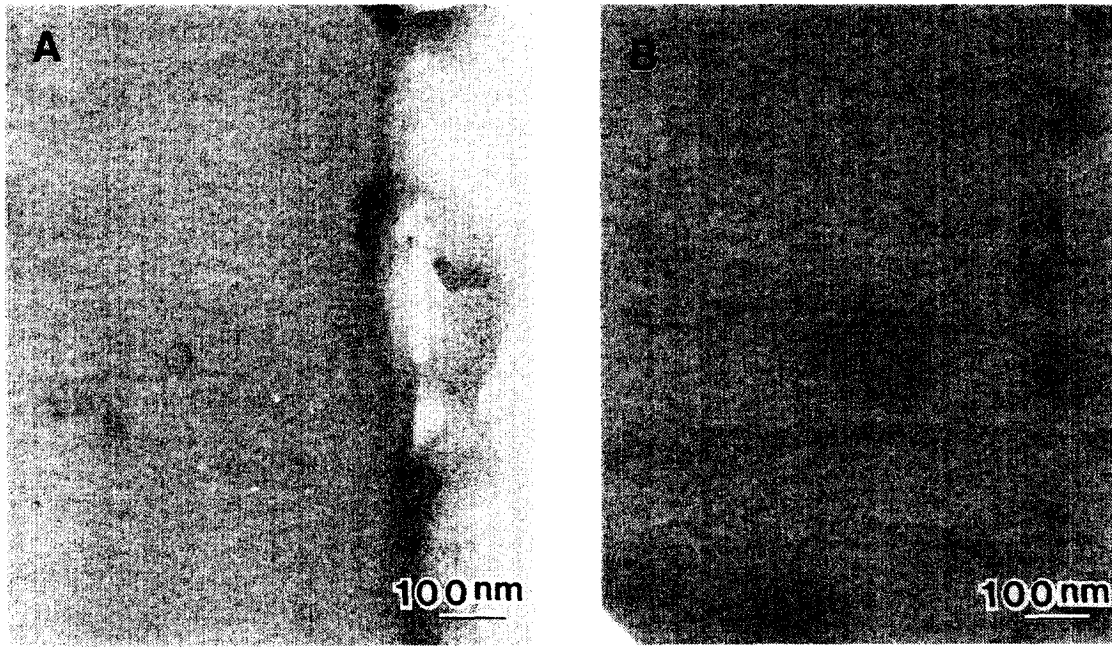

Fig. 8. Electron micrographs of cellulose II.
Notes: (A) : Outer part of a fiber wall. (B) : Inner part of a fiber wall.

presence of regenerated cellulose I in both inner and outer walls and the amount of regenerated cellulose I was rich in inner wall. However, in the case of washing at 0℃, the diagrams show only cellulose II(Fig. 4).

In the previous paper, Kim *et al.* (1990) reported that cellulose I was rich in the internal part, whereas cellulose II was in the external part. They suggested also the heterogeneous localization of reversible and irreversible parts in the fiber wall and a gradient of swelling. Our previous and present works showed that the metastable Na-cellulose I was formed in the inner part by constrained swelling and it could be reconverted into cellulose I by washing with hot water. It was transformed into Na-cellulose IV with washing at low temperature(0℃). Therefore, it is considered that hydration occurs by washing at 0℃, whereas dehydration occurs by washing at 100℃.

3.3 Polarized and electron microscopy

Fig. 6 shows the polarized micrographs of cellulose I(A), the mixture of cellulose I and cellulose II(B) and cellulose II(C) of ramie fibers. These micrographs did not show any differences in microfibril orientation.

The electron micrographs of ultrathin longitudinal sections shows the microfibrils of cellulose I(Fig. 7) and cellulose II(Fig. 8) oriented straightly along with the fiber axis. The difference of microfibrils orientation in the internal and the external layers was not observed. Therefore, it was considered that the disorder of microfibrils caused by mercerization might not occur. However, further investigation would be required to confirm this question, using the methods such as an optical fourier transform method of imaging analysis.

4. CONCLUSION

The crystal transformation of cellulose I to cellulose II was studied by diffraction and microscopic techniques.

X-ray diffraction analysis of samples washed with water at the higher temperature revealed a partial reconversion of Na-cellulose I into cellulose I. The electron diffraction diagrams of regenerated cellulose at 100℃ showed cellulose I in both the inner and the outer layers, and the ratio of cellulose I was higher in the inner layer. The present work suggested that the metastable Na-cellulose I caused by incomplete swelling was present in the inner layer and was transformed into Na-cellulose IV at low temperature washing(hydration) while it was reconverted to cellulose I at high temperature washing(dehydration). Consequently, Na-cellulose I can not be reconverted to cellulose I, and the disorder of microfibrils caused by mercerization might not occur in the fiber wall.

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REFERENCES

1. Fujii, T., K. Shimizu and A. Yamaguchi. 1987. Enzymatic saccharification on ultrathin sections and ultraviolet spectra of Japanese hardwoods and softwoods. *Mokuzai Gakkaishi* 33 : 400~407
2. Hayashi, J., T. Yamada and S. Watanabe. 1974. On the existence of Na-cellulose I_1 and I_{II} and their relation with crystalline structure of original cellulose. *Sen-I Gakkaishi* 30 : T-190~198
3. Hayashi, J., T. Yamada and K. Kimura. 1976. The change of the chain conformation of cellulose from type I to II. *J. Appl. Polm. Symp.* 28 : 713~727
4. Kim, N.H., J. Sugiyama and T. Okano.

1990. X-ray and electron diffraction study of Na-cellulose I. ***Mokuzai Gakkaishi*** 36 : 120~125

5. Nishimura, H., A. Sarko. 1987. Mercerization of cellulose. IV. Mechanism of mercerization and crystallite sizes. ***J. Appl. Polym. Sci.*** 33 : 867~874
6. Okano, T. and A. Sarko. 1985. Mercerization of cellulose. II. Alkali-cellulose intermediates and a possible mercerization mechanism. ***J. Appl. Polym. Sci.*** 30 : 325~332
7. Sisson, W.A. and W. R. Saner. 1941. The effect of the temperature and the concentration of sodium hydroxide on the X-ray diffraction behavior of raw and of degraded cotton. ***J. Phys. Chem.*** 45 : 717~730
8. Spurr, A.R. 1969. A low-viscosity epoxy resin embedding medium for electron microscopy. ***J. Ultrastr. Res.*** 26 : 31~43