NOTE

Relationship Between Cellulase Treatment and Direct Dye Dyeing for Cotton

INTRODUCTION

In order to utilize cellulase in the finishing of cellulose materials such as cotton fabric, the effect of cellulase treatment on the dyeability of the materials must be studied. However, systematic studies cannot be found in this field. Most recently, the cellulose which was previously dyed with reactive dyes, was hydrolyzed by cellulase for analytical purpose.[1]

Cellulase includes different types of enzyme such as exo- and endo-β-glucoxidases.[2] They show different mechanisms of hydrolysis[2] and their roles were discussed in connection with the microstructure of cellulose.[2]

Cellulase is a polymer. Therefore, there is one possibility that hydrolysis by cellulase could occur only in the disordered region of cellulose, due to steric hindrance. In addition, it is interesting to evaluate the extent of hydrolysis of cellulose, which was previously dyed with direct dyes, by cellulase.

From these points of view, cotton fabrics treated with cellulase were dyed with Congo Red and also cotton fabrics dyed with several direct dyes were hydrolyzed with cellulase.

EXPERIMENTAL

Materials

Cotton fabric used was 0.021 cm thick and weighed 108 g/m^2. Counts of warp and weft for the fabric were 19 and 18 tex, respectively. Threads per cm for the fabric were 32 in ends and 27 in picks, respectively. The fabric was purified with aqueous solution of 0.2% nonionic surfactant at 40°C before cellulase treatment.

Congo Red (C.I. Direct Red 28) used was of specific reagent grade. Chrysophenine (C.I. Direct Yellow 11) and Chrysamine (C.I. Direct Yellow 1) were purified by the Robinson–Mills method.[3]

Commercial cellulase (Meicelase) was kindly given by Meiji Seika Co. Ltd. This is from *Trichoderma viride* and was used without further purification. Carboxymethylcellulase activity was 233,000 units/g as determined by the Nelson–Somogyi method.[4] One unit of carboxymethylcellulase was defined as the amount of enzyme which liberate 10 μg of glucose equivalents per 10 minutes.

Cellulase Treatment

About 0.2 g of the samples was treated with an aqueous solution of 0.2% cellulase under the condition of pH 4.5, 40°C, and a liquor to sample ratio of 1:100 for a given time. After the treatment, the cellulase was inactivated in the boiling water. The samples were then neutralized with 0.03% ammonium aqueous solution, thoroughly washed with deionized and distilled water, and dried.

Weight Loss Measurements

Weight loss (WL) was calculated from the following equation:

$$\mathrm{WL}(\%) = \frac{W - W'}{W} \times 100 \qquad (1)$$

where W and W' are weights of the samples before and after cellulase treatment, respectively. The samples were dried at 85°C for 24 h before measurement.

Dyeing Method

About 0.2 g of the fabrics were dyed to equilibrium under the conditions of a liquor to sample ratio of 1 : 100 and 0.05N of NaCl. Dyeing temperatures were in the range of 70–90°C. Dyeing periods were 24, 72, and 120 h at 70, 80, and 90°C, respectively.

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Calculation Method of Affinity of Dyeing $-\Delta\mu^0$

$-\Delta\mu^0$ of the dye was represented by the following equation:[5]

$$-\Delta\mu^0 = RT \ln \frac{[\mathrm{D}]\phi[\mathrm{Na}]\phi^z}{[\mathrm{D}]\sigma[\mathrm{Na}]\sigma^z V^{z+1}} \tag{2}$$

where $[\mathrm{D}]\phi$ = dye uptake of the fiber (g ion/kg), which was obtained here using colorimetric analysis of residual solution, $[\mathrm{Na}]\phi$ = Na^+ concentration in the fiber (g ion/kg), $[\mathrm{D}]\sigma$ = dye concentration in residual solution (g ion/L), $[\mathrm{Na}]\sigma$ = Na^+ concentration in residual solution (g ion/L), z = number of Na in a dye molecule, V = effective volume term of molecules of dyeing for the fiber, R = gas constant 1.99×10^{-3} cal/deg, and T = absolute temperature.

$[\mathrm{Na}]\phi$ was calculated by

$$[\mathrm{Na}]\phi = V[\mathrm{Na}]_i \tag{3}$$

$$[\mathrm{Na}]_i = \frac{[\mathrm{D}]\phi}{V}\left\{\frac{Z}{2} + \left(\frac{Z^2}{4} + \frac{[\mathrm{Na}]\sigma[\mathrm{Cl}]\sigma V^2}{[\mathrm{D}]\phi^2}\right)^{1/2}\right\} \tag{4}$$

where $[\mathrm{Na}]_i$ is the Na^+ concentration in the effective volume of the fiber.

RESULTS AND DISCUSSION

The Change in Dye Uptake by Preceding Treatment with Cellulase

WL of the fabrics increased linearly with an increase in treating time with cellulase, at least, up to 24 h. WL obtained by the treatment for 4 and 24 h was 4.2 and 17.7%, respectively. Untreated fabric and cellulase treated fabrics with WL above were dyed to equilibrium with Congo Red at 90°C and at the initial dye concentration of 4×10^{-4} g ion/L. We attempted first to calculate apparent values of $-\Delta\mu^0$ for these fabrics; from eq. (2) using a constant value 0.22 L/kg of V.[6,7] $-\Delta\mu^0$ calculated were 8.15, 8.06, and 8.25 kcal/mol for 0% (untreated), 4.2%, and 17.7% of WL, respectively.

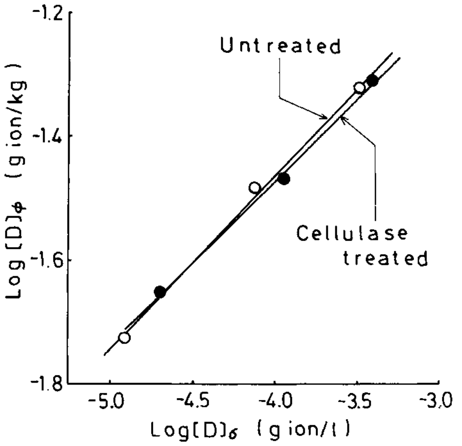

Figure 1 Dyeing isotherms at 90°C for untreated fabric and cellulase-treated fabric with 17.7% of weight loss. $[\mathrm{D}]\phi$ and $[\mathrm{D}]\sigma$ are equilibrium dye uptake of fiber and dye concentration in residual solution, respectively.

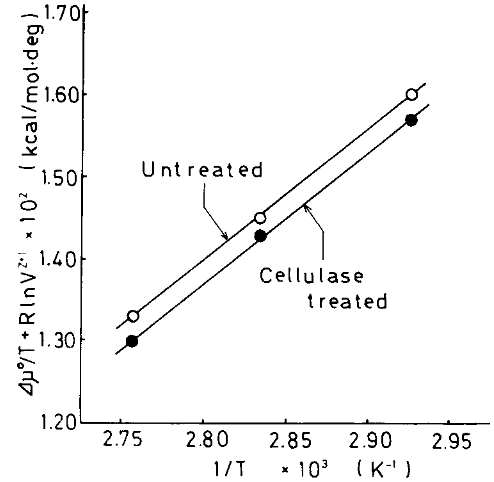

Figure 2 $(-\Delta\mu^0/T + R \ln V^{z+1})$ plots against $1/T$ for untreated fabric and cellulase-treated fabric with 17.7% weight loss. Abbreviations are included in this article.

It is clear that the microstructure of cellulose changes by cellulase-catalyzed hydrolytic cleavage. If the cleavage causes a change in $-\Delta\mu^0$, $-\Delta\mu^0$ would monotonously change in the same direction with increasing WL. However, $-\Delta\mu^0$ obtained above is not consistent with this assumption. It can be assumed that the change in V accompanied by cellulase treatment would play an important role in the change in apparent value of $-\Delta\mu^0$ calculated above.

If $-\Delta\mu^0$ is approximately constant regardless of cellulase treatment and a suitable value is taken as V_0, the relative value of V_e/V_0 can be obtained using eq. (2) by numerical calculation, where V_0 and V_e are V's for untreated and cellulase-treated fabrics, respectively.

Dyeing isotherms at 90°C were obtained for untreated fabric and cellulase-treated fabric having 17.7% of WL. Linear relationship between log $[\mathrm{D}]\phi$ and log $[\mathrm{D}]\sigma$ can be observed for each fabric, as shown in Figure 1. Isotherms obey Freundlich type[8] regardless of the treatment. Two slopes observed here were nearly 0.3. It was suggested that the dye adsorption mechanism might be similar for untreated and cellulase-treated fabrics.

The change in enthalpy of dyeing would give one of the most important criteria[9] to estimate the change in the nature of the interaction between dye molecule and binding site of cellulose, caused by cellulase treatment. $(-\Delta\mu^0/T + R \ln V^{z+1})$ can be plotted against $1/T$ from eq. (2).

The plots for untreated fabric and cellulase-treated fabric with 17.7% WL are shown in Figure 2. The linear line can be observed for each fabric, and the two slopes observed, which are enthalpy of dyeing, are nearly equal.

The above experimental results regarding dyeing isotherm and enthalpy of dyeing are coincident with the assumption that $-\Delta\mu^0$ may be nearly equal for cellulase-treated and untreated fabrics.

The fabrics having various values of WL were dyed with Congo Red at 90°C and at an initial dye concentration of 4×10^{-4} g ion/L, and then V_e/V_0 was obtained for each sample. Calculation of V_e/V_0 was done using 0.22 L/kg of V_0, on the assumption that $-\Delta\mu^0$ may be constant regardless of the extent of hydrolysis. V_e/V_0 plots against WL are shown in Figure 3. It is noteworthy that V_e/V_0 is smaller than 1 at smaller WL. V_e/V_0 shows minimum in the curve and then gradually increases with WL. V_e/V_0 is greater than 1 at a considerably larger value of WL. From these results, it can be assumed that cellulase-catalyzed hydrolysis would occur mainly in the amorphous region at smaller WL. It can be also assumed that the crystalline region may be hydrolyzed gradually from its surface and would be partially transformed into amorphous region, by cellulase treatment.

Weight Loss of the Dyed Fabrics by Hydrolysis

The fabrics, which were previously dyed to equilibrium with Congo Red, Chrysamine and Chrysophenine, were treated with cellulase for 24 h and then the WL was examined. The dyeing was done at 80°C in the range of the initial dye concentrations of 0.748×10^{-4}–2.99×10^{-3} g ion/L for Chrysamine, 1×10^{-4}–4×10^{-4} g ion/L for Chrysophenine, and 1×10^{-4}–8×10^{-4} g ion/L for Congo Red. The plots of WL against dye uptake are shown in Figure 4. WL tends to decrease with increasing dye uptake for all of three dyes used. It is clear that WL of the fabrics dyed are smaller in the order of Congo Red, Chrysamine, and Chrysophenine at a given dye uptake. $-\Delta\mu^0$ obtained at 80°C using 0.22 L/kg of V_0 was 8.33, 5.52, and 4.85 kcal/mol for Congo Red, Chrysamine, and Chrysophenine, respectively. Therefore, it was concluded that cellulose molecules bound by the dyes, the affinity of which is larger, could be attacked by cellulase with more difficulty.

The chemical structure of cellulose does not change by dyeing with direct dyes. It was considered that physical interactions such as hydrogen bonding, which was formed between the direct dye and the binding site, may block the hydrolysis of cellulose by cellulase, to the extent associated with the affinity of the dyes.

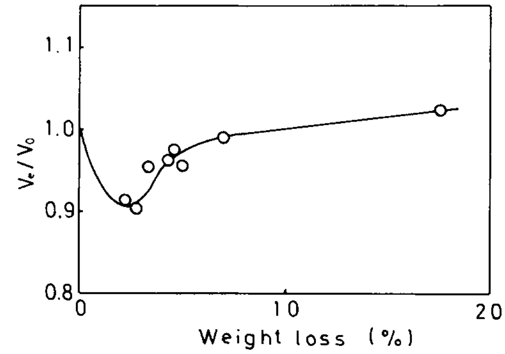

Figure 3 V_e/V_0 plots against weight loss of cotton fabrics caused by cellulase treatment. V_0 and V_e are effective volume terms of dyeing for untreated and cellulase-treated fabrics, respectively.

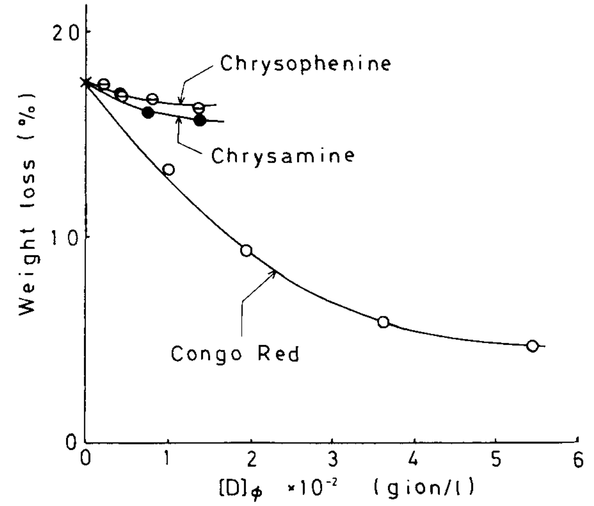

Figure 4 Relationship between weight loss caused by cellulase treatment and equilibrium dye uptake of fiber $[D]\phi$. The fabrics were dyed with the dyes indicated and then hydrolyzed by cellulase.

CONCLUSION

Equilibrium dye uptake of Congo Red by cotton fabrics, which was previously treated with cellulase, was measured, and then the relative volume term of dyeing was obtained in connection with weight loss. As a result, it was assumed that cellulase-catalyzed hydrolysis would occur mainly in the amorphous region at smaller WL. Besides, the fabrics previously dyed to equilibrium with several direct dyes were treated with cellulase. WL by hydrolysis tended to decrease with increasing dye uptake in every case of dye. Also, WL decreased with increasing affinity of the dye for cellulose, at a given dye uptake.

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