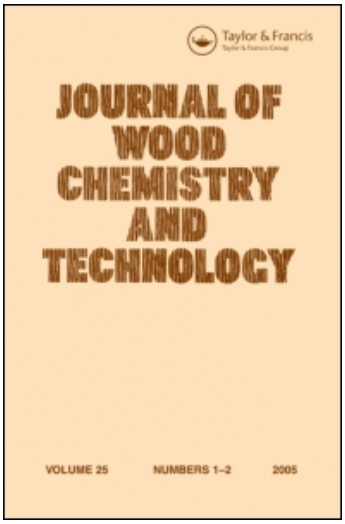

Journal of Wood Chemistry and Technology
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597282

Mechanism of Dilute Acid Hydrolysis of Cellulose Accounting for its Degradation in the Solid State

Jean Bouchard; Ralph P. Overend; Esteban Chornet; Marie-Rose Van Calsteren

To cite this Article Bouchard, Jean , Overend, Ralph P. , Chornet, Esteban and Van Calsteren, Marie-Rose(1992) 'Mechanism of Dilute Acid Hydrolysis of Cellulose Accounting for its Degradation in the Solid State', Journal of Wood Chemistry and Technology, 12: 3, 335 — 354
To link to this Article: DOI: 10.1080/02773819208545239
URL: http://dx.doi.org/10.1080/02773819208545239

JOURNAL OF WOOD CHEMISTRY AND TECHNOLOGY, 12(3), 335-354 (1992)

MECHANISM OF DILUTE ACID HYDROLYSIS OF CELLULOSE ACCOUNTING FOR ITS DEGRADATION IN THE SOLID STATE

Jean Bouchard[1], Ralph P. Overend[2] and Esteban Chornet
Chemical Engineering Department, University of Sherbrooke
Sherbrooke, Québec, Canada J1K 2R1

Marie-Rose Van Calsteren
Food Research and Development Centre of Canada
St-Hyacinthe, Québec, Canada J2S 8E3

ABSTRACT

It is known that dilute acid hydrolysis of cellulose at high temperature results in yields not exceeding 60-65% of the potential glucose. All the models presented in the literature are based on the sequence: cellulose → glucose → degradation products. The underlying assumption of this sequence is that the unconverted cellulose retains its chemical integrity during the course of the hydrolysis reaction. Experimental evidence based on physico-chemical characterization of cellulosic residues obtained from dilute acid hydrolysis processes allows us to propose a new mechanism for cellulose hydrolysis accounting for its degradation in the solid state. Such degradation results in the formation of a non-carbohydrate residue.

---

1. Present address: Pulp and Paper Research Institute of Canada, 570 Boul. St-Jean, Pointe Claire, Québec, Canada, H9R 3J9

2. Present address: National Renewable Energy Laboratory, 1617 Cole Boul., Golden, CO 80401

INTRODUCTION

Hydrolysis of cellulose to glucose using concentrated inorganic acid followed by a milder secondary hydrolysis yields 95% of the glucose potential of cellulose. However, hydrolysis by dilute inorganic acid at elevated temperatures results in yields limited to 60-65% of the potential glucose. Many chemical models have been proposed to explain these yields[1-3], most of them being variations of Saeman's kinetic model. In 1945, he was the first to propose the following sequence:

Cellulose -----> Glucose -----> Degradation products

None of the models considers the possibility of physico-chemical transformations of the cellulosic substrate itself during the course of this reaction as being partially responsible for the observed limited glucose yields. The recent model for dilute acid hydrolysis of cellulose, proposed by Conner *et al.*[3], incorporates the reversion and degradation reactions of glucose and the presence of readily hydrolysable cellulose. In this model, the chemical transformation of glucose (i.e. degradation) appears to be the main reason for the low sugar yield observed in dilute hydrolytic processes. Acid hydrolysis of cellulose has also been studied in order to relate the kinetics of depolymerization to the variation in the molecular weight distribution[4-6] but very little attention has been paid so far to the characteristics of the cellulosic residues obtained via partial dilute acid hydrolysis as the reaction proceeds[7-9].

We have recently demonstrated that dilute acid hydrolysis of cellulose at elevated temperatures involves

a chemical transformation of a fraction of the cellulose in the polymeric state[10]. This chemically modified cellulose cannot yield 100% glucose by further hydrolytic treatment and is partially responsible for the low saccharification yields obtained from these processes. Such an effect has led to the consideration that a new pathway for cellulose degradation needs to be introduced into the overall mechanism for the prediction of conversion and sugar yields from dilute acid hydrolysis of cellulose. By comparing dilute acid hydrolysis at low and high temperatures as a function of the time of the reaction, we have previously shown[10], that the extent of depolymerization as a function of the conversion to solubles were identical for both processes. Chemical characterization of cellulosic residues indicates that significant degradation of the cellulose takes place in the solid state. This degradation in solid state is directly responsible for a decrease of 10% of the glucose yield at low temperature (100°C) while a 35% decrease of yield is observed for hydrolysis at high temperature (190°C). Cellulose degradation is the unique responsible for this decrease in yield which should be added to the one cause by glucose degradation pathway to account for the overall yield. FTIR studies have indicated that the chemical characteristics of the degraded cellulose are independent of the process used while the extent of degradation is a function of the severity of the treatment. Acid-catalysed degradation of cellulose in the solid state implies formation of carbonyl groups and, possibly, carbon-carbon double bonds formed by dehydration.

The objective of the present paper is to complete the cellulose acid hydrolysis mechanism by: providing

experimental evidence of the extent of solid cellulose degradation in a continuous acid hydrolysis process as a function of the temperature of the reaction; gaining information on the nature of structural and chemical degradation reactions along the hydrolytic sequences.

RESULTS AND DISCUSSION

Two series of experiments have been carried out as described in the Experimental Section. A standard 100°C analytical hydrolysis using dilute hydrochloric acid (H-An series) serves as a reference for the higher severity continuous process (H-Co series). Table 1 shows the conversion obtained and the sugar potential of the residual solid cellulose as a function of the time of hydrolysis for H-An Series, and of the temperature of the reaction for H-Co Series at two acid concentrations.

The results from the low-temperature hydrolysis (H-An Series) indicate that the integrity of the solid cellulose is preserved during half of the hydrolysis reaction (50% of conversion). Thereafter the hydrolysis is accompanied by degradation of the solid cellulose and consequently by a decrease of the glucose potential of the remaining cellulose. Pursuing the hydrolysis to high conversion (85%) leads to significant degradation of the cellulose where glucose accounts only for 30% of its chemical composition. Acid hydrolysis at higher temperatures (H-Co Series) leads also in degradation of the remaining cellulose. Due to the effect of high temperature, the solid state degradation of cellulose can be observed as only 20% of it has been solubilized as compared to 50% in the case of the reference hydrolysis

TABLE 1

Conversion to soluble products and glucose potential of the residues obtained from the hydrolysis of cellulose by standard (H-An) and continuous (H-Co) processes where number 1 and 2 refer to experiments done with sulfuric acid at concentrations of 0.05 M and 0.12 M respectively.

Sample	Conversion (%)	Glucose Potential of Residue (%)
H-An Series		
0 h	1.0	99.8
1 h	17.5	99.5
2 h	31.5	95.3
4 h	37.5	95.3
7 h	51.9	95.6
16 h	68.7	74.7
24 h	78.5	70.2
48 h	87.4	30.6
H-Co Series		
1-200°C	24.3	83.6
1-220°C	46.8	69.4
1-230°C	80.5	63.0
1-240°C	94.4	18.8
2-200°C	71.0	36.7
2-220°C	96.6	16.7
2-230°C	95.8	1.3
2-240°C	94.1	0

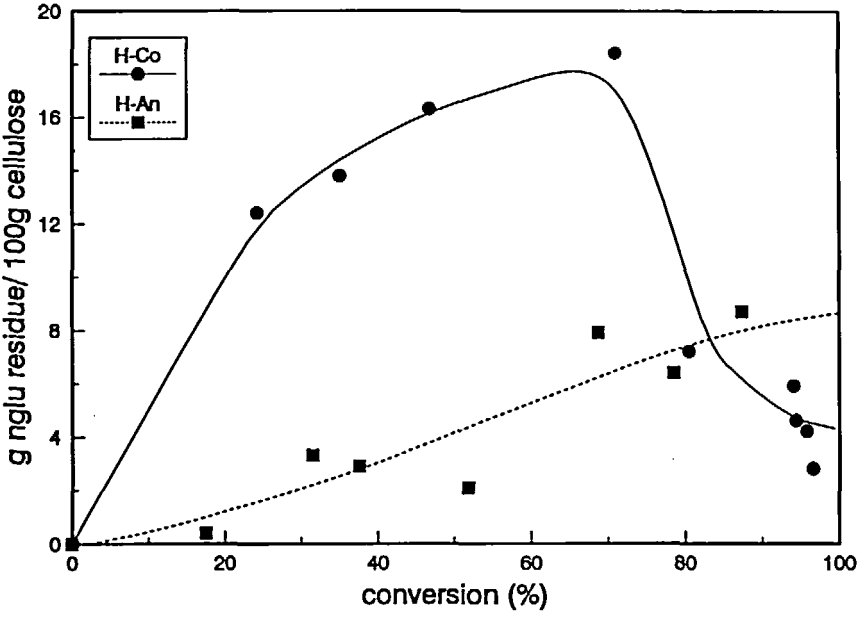

FIGURE 1: Non-glucosidic structure formation (in g/100g of initial cellulose) as a function of conversion for H-An and H-Co series

(H-An). These results mean that for a 40% conversion, the remaining residu (60g) is anhydroglucose for the low-temperature H-An hydrolysis while it is composed of 44 g of anhydroglucose and 16 g of unknown for the high-temperature hydrolysis.

From now, the degraded part of the cellulose will be identified as non-glucosidic residue or "n-glu". The concentrations of n-glu (in g/100g of initial cellulose) are plotted as a function of the conversion for both series in Figure 1.

In the case of the H-An series, the concentration of n-glu increases almost linearly as a function of the

conversion and reaches a maximum value of 8g/100g of cellulose. However, a different behavior is observed for the high temperature series (H-Co). The presence of n-glu appears at low conversions and reaches a maximum value of 19g/100g of cellulose at 70% of conversion. At higher conversions, the concentration of n-glu has strongly decrease indicating that an important part of the n-glu residue has gone into solution as soluble degradation products. Performing the hydrolysis to the limit leads to the formation of a final non-glucosidic residue representing 4-5% of the initial cellulose. The lower formation of n-glu in solid cellulose for the H-An hydrolysis can be attributed to a lower temperature of reaction or to the fact that the higher acid concentration (2.5N) is kinetically more favorable to hydrolysis as compare to solid state degradation.

The molecular weight distribution of the cellulosic residues has been obtained by size exclusion chromatography of their carbanilate derivatives. Figure 2 shows the chromatograms of initial cellulose and the residues of the H-An series (identified by the hydrolysis time). From these chromatograms and those of the H-Co series which are not shown, a similar evolution of their molecular weight distributions can be described.

A rapid depolymerization as the amorphous regions are hydrolysed. This is followed by the level-off degree of polymerization step, LODP, where the form of the distribution is maintained as the crystallites can only be hydrolyzed from their surfaces and ends[11]. At the end of the LODP step, we observe the formation of a bimodal distribution caused by the appearance of a low molecular weight fraction. We have shown previously that degradation of cellulose is becoming important at this point

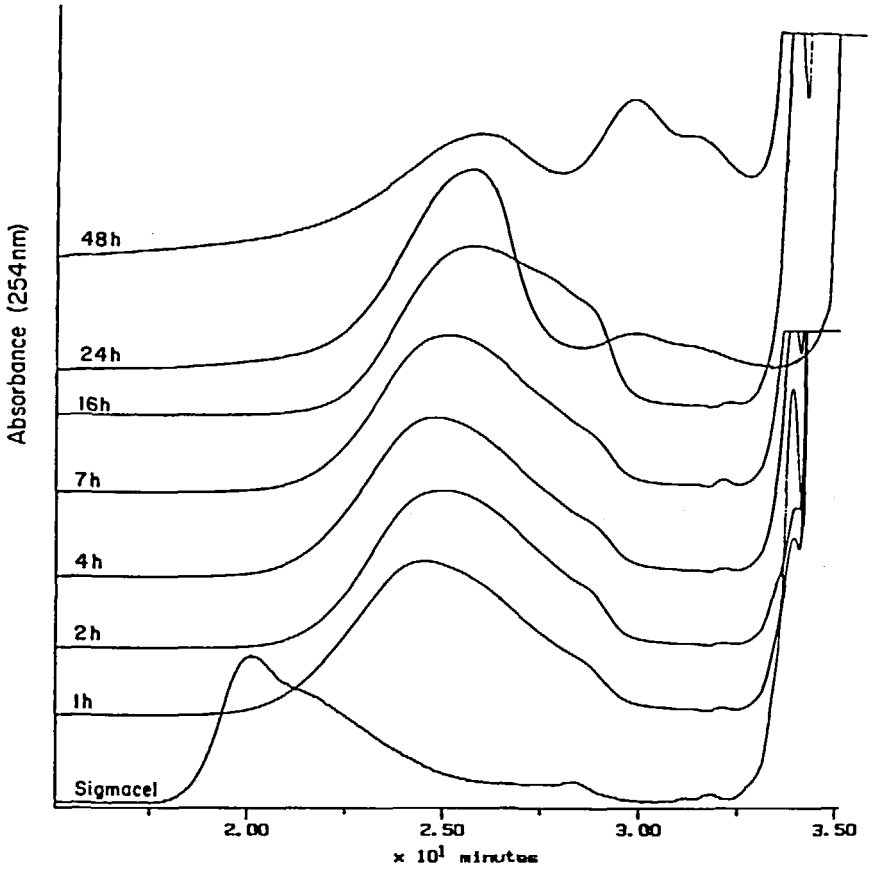

FIGURE 2: Size exclusion chromatograms of cellulosic residues of H-An series

TABLE 2

Crystallinity indexes as calculated from X-ray diffraction (CrI) and ^{13}C CP/MAS NMR ($C\text{-}4_{Cr}/C\text{-}4_{Am}$) of cellulosic residues

Sample	DP_W	CrI (%) (X-ray)	$C\text{-}4_{Cr}/C\text{-}4_{Am}$ (NMR)
Sigmacel	1220	78.4	1.3
H-An Series			
1 h	172	85.9	---
2 h	141	87.1	---
4 h	124	86.2	2.1
7 h	116	86.0	2.3
16 h	77	82.2	---
24 h	78	79.0	2.5
48 h	66	67.5	2.4
H-Co Series			
1-200	150	94.1	---
1-220	105	90.1	---
1-230	81	88.9	1.9
2-200	---	81.7	---
1-240	21	63.7	2.1
2-220	---	53.8	---
2-230	---	0	---

leading to non-glucosidic structures. The hypothesis of formation and accumulation of insoluble oligosaccharides (DP 5-10) can be rejected because they should be rapidly hydrolysed and solubilized. The results suggest rather the formation of insoluble and degraded residues with a "carbanilated" molecular weight ranging between 3000 and 5000. However, because we don't know the degree of

carbanilation of this specific fraction, we cannot state on its real molecular weight.

Table 2 shows the crystallinity indexes for the cellulosic residues of both series, suggesting that the evolution of these parameters is similar for both processes. From an initial value of 78%, the crystallinity index increases to a maximum at the LODP. Continuation of the hydrolysis leads to a decrease of the index and finally to formation of an amorphous non-glucosidic residue. However, it can be seen that the acid hydrolysis using the continuous process (H-Co) results, for similar conversions, in the production of cellulose with higher crystallinity. The cellulose obtained by this process also keeps its crystalline properties at a lower degree of polymerization.

Zhbankov *et al.*[12] have proposed that the ratio of the carbon 4 peaks ($C\text{-}4_{Cr}/C\text{-}4_{Am}$) from the ^{13}C CP/MAS NMR spectra respectively found at 89 and 84 ppm of chemical shift, can be used as an index of order and related to the crystallinity. Furthermore, from experiments on bleached kraft pulp, Newnam *et al.*[13] have shown that the $T_{1\rho}$ for crystalline and non-crystalline cellulose are indistinguishable. As it can be seen in Table 2, both crystallinity indexes, from X-ray diffraction and NMR, increase in the first stage of the hydrolysis as the amorphous regions are hydrolysed. Beyond this step, hydrolysis and degradation of the crystallites occur simultaneously at their surfaces. Because X-ray diffraction measures the concentration of the ordered region in the whole sample, the decrease of the crystallinity index is correlated with the decrease of the cellulose concentration in the solid residue.

The NMR index is unaffected by degradation because it is specifically related to the carbon 4 of the intact anhydroglucose units in the residue. This means, as an example, that for the H-An (48h) sample, the NMR index is related to the crystallinity of the 30% of the sample which is cellulosic without any interference from the 70% of non-glucosidic part. Consequently, this particular index should stay stable except if decrystallisation occurs within the crystallites. The fact that this index is not affected while the X-ray index decreases is an indication that as the surface is solubilized and/or degraded, the undegraded cellulose remains highly crystalline. The same trend can also be observed from the signal of the C-6 carbon (66 and 63 ppm).

Figure 3 and 4 show the solid-state ^{13}C CP/MAS NMR spectra of cellulosic residues from 0 to 250 ppm of chemical shift. The 60-120 ppm area represents the different carbons of the cellulose. The study of this area indicates clearly that part of the cellulose remains undegraded even for low DP samples. The sample 2-230 in Figure 4 shows the absence of cellulosic structure which correlates with the glucose potential of this residue (1.3%). The other areas of the spectra of Figure 3 and 4 suggest the formation of new chemical structures following the acidic degradation of the cellulose. Four chemical structures can be related to the chemical shift of the peaks: carbonyl groups from ketone at 210 ppm; from carboxylic acid at 175 ppm; unsaturated carbons in the 110-160 ppm area with specific peaks at 110 and 150 ppm which can be related to a double bond formed between carbons 5 and 6 of glucose unit; saturated aliphatic carbon in the 10-50 ppm area.

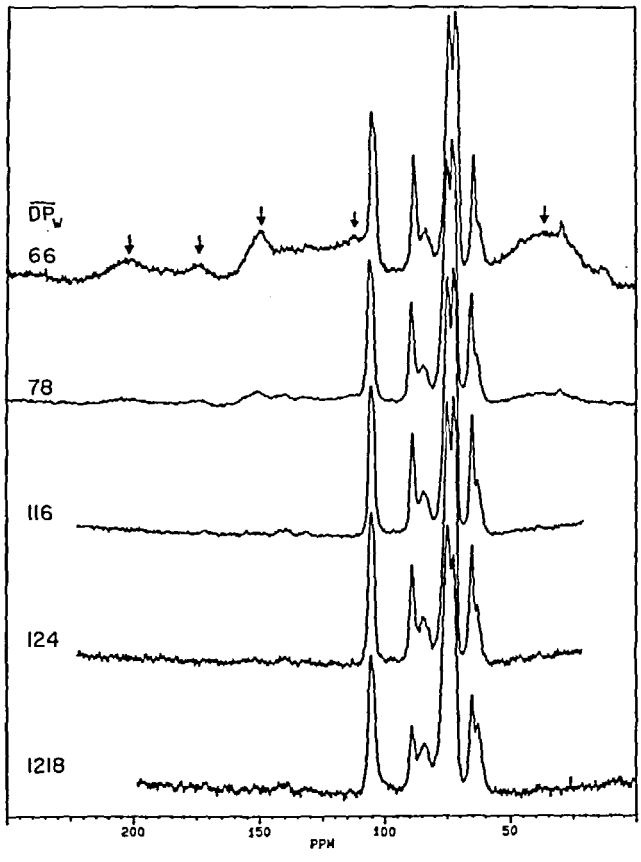

FIGURE 3: ^{13}C NMR (CP/MAS) spectra of cellulosic residues of H-An series (0-250 ppm)

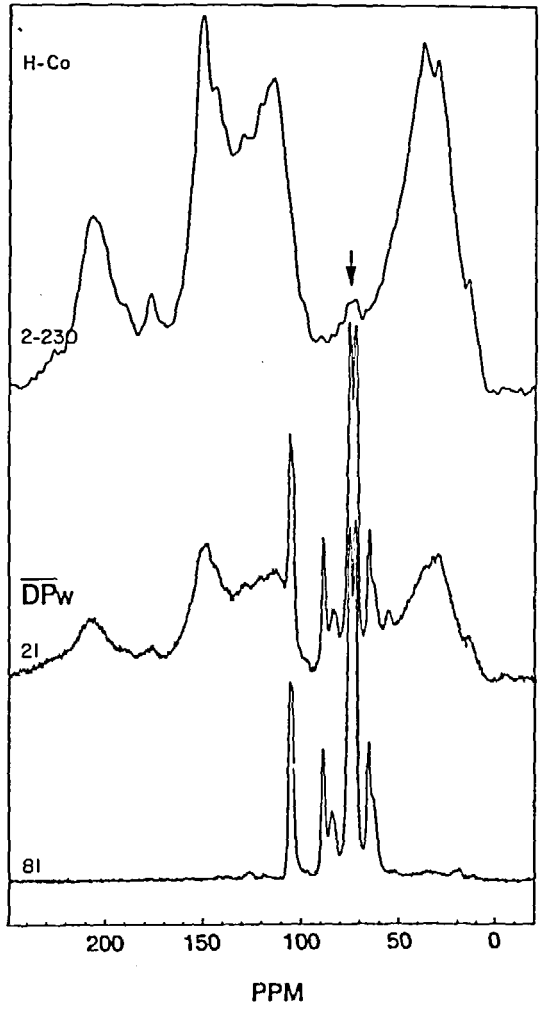

FIGURE 4: ^{13}C NMR (CP/MAS) spectra of cellulosic residues of H-Co series (0-250 ppm)

Discussion on a Chemical Model for Dilute Acid Hydrolysis of Cellulose

When cellulose is hydrolysed with dilute inorganic acid, à chemical degradation of the solid cellulose occurs as the depolymerization and the solubilization proceed. This degradation begins during the LODP step and becomes more important thereafter. The principal effect of this degradation is a decrease of the glucose potential of the cellulosic residue and consequently of the overall saccharification process.

The existing models for cellulose hydrolysis attribute the low glucose yield only to the reversion and degradation reactions of glucose once in solution. We have demonstrated tha the degradation of the cellulose in the solid state also occurs and propose the mechanism shown in Figure 5.

The degradation of the cellulose substrate must proceed via dehydration and oxidation reactions identical for both processes but activated by temperature. This degradation occurs at the surface of the crystallite and the internal highly crystalline cellulose remains intact. Our observation that cellulases can not hydrolyse degraded residues even if they are partially composed of anhydroglucose suggests also a degradation of the surface. Formation of ketonic groups on carbons 2 and 3 and/or carboxylic acids on carbon 6, and possibly on carbons 2 and 3 after ring opening are suggested from FTIR[10] and NMR results. Furthermore, dehydration reactions leading to double bonds formation on C_2-C_3 and C_5-C_6 can be proposed from the NMR data. The degradation results in the formation of a non-glucosidic structure in the cellulosic residue which is partially soluble. The

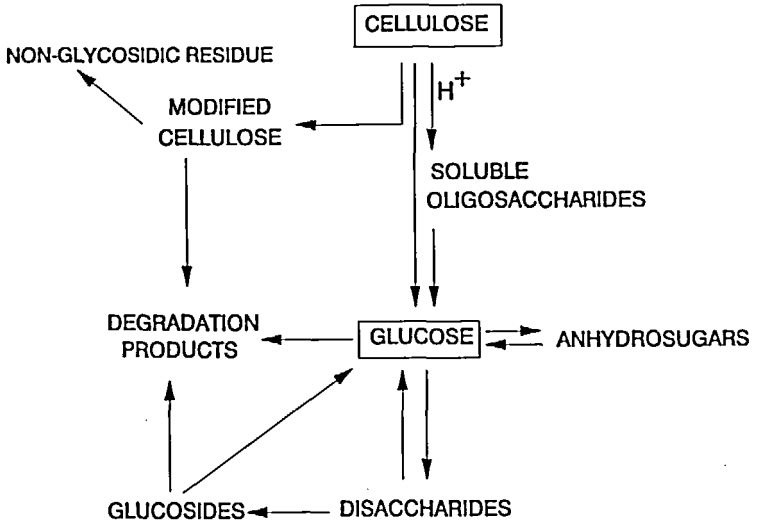

FIGURE 5: Model for dilute acid hydrolysis of cellulose accounting for its degradation in the solid state

final n-glu residue has a molecular weight in the 3000-5000 range after carbanilation and could be highly cross-linked and partially aromatic.

CONCLUSIONS

During acid hydrolysis, the glucose potential of the remaining cellulose decreases with the extent of depolymerization. This phenomenon is enhanced by conducting the hydrolysis at high temperature. This pathway of solid state degradation should be introduced into the proposed mechanism of cellulose hydrolysis.

The degradation reactions involve formation of carbonyl groups from both ketone and carboxylic acid and

also carbon-carbon double bonds from dehydration reactions. Degradation finally results in the formation of a non-glucosidic macromolecule representing about 5% of the initial cellulose.

Although the overall crystalline property order decreases during the degradation, the intact cellulose, probably located in the center of the microfibril, appears to be highly ordered suggesting that acid hydrolysis from the surface does not create defects inside the crystallites.

EXPERIMENTAL

Analytical Hydrolytic Approach (H-An series)

Batches of 5 g of cellulose (Sigmacel No. C-8002, Sigma Chem. Co.) were hydrolysed separately in 250 mL round-bottomed flasks under reflux (103-105°C) and inert atmosphere (nitrogen) in a solution of 2.5 N hydrochloric acid. These conditions are identical to those used in others studies[4,7,9,11]. The concentration of cellulose was 5% w/w and the reaction time varied between 0 and 48 hours. The reaction was stopped by cooling in iced water and the solution was filtered under vacuum with a Buchner funnel. The cellulosic residue was washed with distilled water until neutrality, dried at 60°C and weighed.

Continuous Hydrolysis Process (H-Co series)

Two series of hydrolyses (H-Co series) have been conducted in a continuous hydrolysis unit developped in 1985 and combining a soaking recirculation section and a tubular reactor[14]. In the soaking section, the cellulose

suspension (6%) in water is kept recirculating until transferred to a second section of the unit where it is rapidly heated to the desired temperature (200-240°C) by steam injection. After passage of the heated suspension through a homogeneizing valve, sulfuric acid is added at concentration of 0.05 M or 0.12 M (series 1 and 2). The hydrolysis occurs then in a tubular reactor for 2 minutes. Before flashing the suspension, a solution of sodium hydroxide is injected in order to adjust the pH to about 5. The suspension is filtered and the solid cake is washed with water then dried at 60°C.

Analytical Methodology

For each experiment, the conversion representing the percentage of initial material that has been solubilized was calculated. The conversion does not correspond to the glucose production but rather to the total yield of soluble molecules irrespective of whether further reactions take place in solution. The procedures for determination of the sugar potential of the cellulosic residues have been described previously[10].

The molecular weight distribution of the cellulosic residues was determined by size exclusion chromatography of their tricarbanilate derivatives. Carbanilation is conducted following the procedure proposed by Wood *et al.*[15]. The SEC was performed using four columns connected in series (PLgel, 300 x 7.5mm, Polymer Lab.) of pore size of 50, 500, 10^4 and 10^5 Å respectively. UV detection was made at 254 nm. The following operating conditions were applied: eluent, THF (1 mL/min); temperature, 40°C; injection volume, 100 μL; sample concentration, 3-6 mg/mL in THF (filtered on 0.45 μm filter). Determination of molecular weights is made from

universal calibration[16] using polystyrenes as standards and the mean of the Mark-Houwink coefficients proposed in the litterature[17-20] for polystyrene (PS) and cellulose tricarbanilate (CTC): $\alpha_{PS} = 0.699$; $K_{PS} = 0.0158$; $\alpha_{CTC} = 0.857$; $K_{CTC} = 0.0040$.

X-ray diffractograms were recorded using a Rigaku "Geiger Flex" equipped with a copper source (lambda = 1.542 Å) to which a 40 mA was applied at a potential of 30 kV. A nickel filter was used to eliminate the kBeta. Spectra were recorded between 10 and 40° of 2Θ angle at a speed of 3°/min. Crystallinity indexes were determined using the method proposed by Segal *et al.*[21].

The ^{13}C CP/MAS NMR spectra were recorded using a 7 mm double bearing probe on a Bruker AM-300 instrument equipped with MSL high-power amplifiers. The respective frequencies for ^{1}H and ^{13}C were 300.13 and 75.47 MHz. About 500 mg of cellulose sample were pressed in an alumina rotor to which a 5 kHz spinning rate was applied at a temperature of 308°K. The pulse sequence was a single contact optimized to 2 ms. The length of the $\pi/2$ pulse was 6 μs, 8 s of recycle time and detection was made with quadrature. Phase alternation was used throughout the signal acquisition to minimize baseline and intensity artifacts. The magic angle was adjusted by monitoring the ^{79}Br spectrum of KBr. The chemical shifts relative to tetramethylsilane were determined by using the CH peak of adamantane at 38.3 ppm as an external standard. Between 1000 and 5000 scans were co-added.

ACKNOWLEDGMENTS

The authors are indebted to the Bioenergy Development Program of EMR, Canada for financial support. Technical assistances of G. Phaneuf, A. Roireau and T.S. Nguyen are gratefully acknowledged. FRDCC publication number: 242.

REFERENCES

1. J.F. Saeman, Ind. Eng. Chem. 37, 43 (1945).
2. H.E. Grethlein, Biotech. Bioeng. 20, 503 (1978).
3. A.H. Conner, B.F. Wood, C.G. Hill and J.F.Harris, J.Wood Chem. Technol. 5, 461 (1985).
4. R.J. Elema, J. Pol. Sci. Symp. No.42, 1545 (1973).
5. R.L. Fellen, S.B. Lee and J. Bogaard, Adv. Chem. Ser. 212, 329 (1986).
6. M. Marx-Figini, J. Appl. Pol. Sci. 33, 2097 (1987).
7. O.A. Battista, S. Coppick, J.A. Howsmon, F.F. Morehead and W.A. Sisson, Ind. Eng. Chem. 2, 333 (1956).
8. Y.V. Svetkin, V.T. Rovenskii, G.P. Lomova, L.V. Koval, N.M. Malinovskaya, I.S. Palii, S.M. Lisitskaya and O.A. Polonskaya. Gidroliznaya i Lesokhimicheskaya Promyshlennost No. 4, 3 (1985).
9. T. Hattula, Paperi ja Puu 69, 92 (1987).
10. J. Bouchard, N. Abatzoglou, E. Chornet and R.P. Overend, Wood Sci. Technol. 23, 343 (1989).
11. A. Sharples, Trans. Faraday Soc. 53 1003 (1957).
12. R.G. Zhbankov, M.Y. Ioelovich, A. Treimanis, E.T. Lippmaa, R. Teejaer, F.N. Kaputskii, D.D. Grinshpan and L.G. Lushkin, Khim. Drev 3-6 (1986)
13. R.H. Newman and J.A. Hemmingson, Holzforschung 44 351 (1990).
14. P. Koeberle, F. Meloche, G. Chauvette, H. Ménard, E. Chornet, L. Jaulin, M. Heitz and R.P. Overend, In Fifth Canadian Bioenergy R&D Seminar, S. Hasnain (ed.), Elsevier Applied Sciences Publ., 1985, p. 263.
15. B.F. Wood, A.H. Conner and C.G. Hill, J. Appl. Pol. Sci. 32, 3703 (1986).
16. H. Benoit, Z. Grusibic, P. Rempp, D. Decker and J. Zilliox, J. Chim. Phys. 63, 1507 (1966).
17. M. Kolinsky and J. Janca, J. Pol. Sci. Pol. Chem. Ed. 12, 1181 (1974).

18. A.E. El Ashmawy, J. Danhelka and I. Kossler, Svensk Papperstidn. 16, 603 (1974).

19. J.J. Cael, R.E. Cannon and A.O. Diggs, ACS Symp. Series 150, 43 (1980).

20. J. Danhelka, I. Kossler and V. Bohackova, J. Polym. Sci. Pol. Chem. Ed. 14, 287 (1976).

21. L. Segal, J.J. Creely, A.E. Martin and C.M. Conrad, Textile Res. J. 29, 786 (1959).