JOURNAL OF APPLIED POLYMER SCIENCE VOL. 16, PP. 3063-3071 (1972)

Spectroscopic Investigation of Cellulose Degraded Celluloses, and Their Phenylhydrazine Derivatives in Relation to Assessment of Degradation

H. S. BLAIR and R. CROMIE,* *Department of Chemistry (Industrial), Queen's University, Belfast, Northern Ireland*

Synopsis

The ultraviolet absorption spectra of Cadoxen solutions of hydrocellulose and hydrocellulose phenylosazones are described. With solutions of hydrocellulose no simple relationship between D.P. and absorbance was found. Solutions of hydrocellulose phenylosazone in Cadoxen show well-defined maxima at 277 nm and 390 nm and do not change over 1 hr when oxygen is excluded. The absorbance of the phenylosazone at 390 nm may be used to determine the D.P. of the hydrocellulose samples.

INTRODUCTION

The determination of the degree of degradation of cellulose occurring during processing is very important. The major technique used for assessing depolymerization is the fluidity of the material in solution in alkaline copper complex solvents. While these methods are of considerable value, the relationship between fluidity and degree of degradation is empirical, and a spectroscopic method based on solutions of cellulose or its derivatives would be advantageous.

Little has been published on the ultraviolet absorption spectra of cellulose and its derivatives, and this is confused due, in part, to the lack of suitable solvents.

Fibrous crystalline cellulose is soluble only in the presence of strongly solvating agents and is liable to chemical degradation during the process. Acidic solvents are seldom used as the degradation is then hydrolytic and may only be retarded by lowering the temperature.[1] In alkaline solvents, the degradation is oxidative and may be minimized by the exclusion of oxygen, or possibly by the addition of antioxidants.[2]

Cadoxen, first described in 1957 by Jayme and Neuschäffer,[3] is now acknowledged to be one of the most useful solvents for cellulose. It is transparent, colorless and odorless, and is stable for long periods.[1] Cadoxen can dissolve marked amounts of cellulose at room temperature;

* Present address: Northern Ireland Polytechnic, Jordanstown, Co. Antrim, Northern Ireland.

further, it can dissolve high molecular weight cellulose, MW > 10^6, causing negligible degradation of the dissolved polymer.[4]

Cadoxen does not absorb appreciably above 240 nm and so should be valuable as a solvent for studying the ultraviolet absorption spectra of cellulose and its derivatives. Donetzhuber[5] investigated the absorption spectra of solutions of cellulose pulps in Cadoxen and reported a well-defined maximum at 234 nm and that the specific extinction coefficient was the same for all the pulps examined. He suggested that ultraviolet absorption spectroscopy could be used to determine the concentration of cellulose pulps in Cadoxen. Donetzhuber later[6] studied the ultraviolet absorption spectra of solutions of cotton linters in Cadoxen but found no characteristic absorption. Sjöström and Enström[7] investigated the spectroscopic determination of lignin in pulps and reported that the absorption of cellulose in Cadoxen increased rapidly below 250 nm but did not detect any well-defined maximum.

If a suitable solvent is available, it should be possible to determine the molecular weight of a polymer by spectroscopic endgroup analysis. A number of conditions, however, must be fulfilled: (i) The number of endgroups being determined per molecule must be known, and so the method in general is confined to linear polymers. (ii) There must be complete reaction of the specific reagent with the functional endgroup, and side reactions must not occur. As the size of the molecule increases, there may be incomplete reaction of the reagents with the functional group. (iii) As the number of endgroups is inversely proportional to the molecular weight, determination of molecular weight by endgroup analysis is applicable only to polymers with molecular weight less than 50,000, although by use of special techniques it has been used for molecular weights as high as 10^6.[8]

The simple theoretical basis for determination of number-average molecular weight ($\overline{M_n}$) by endgroup analysis depends on the relationship

$$\overline{M_n} = f \cdot w/N$$

where f is the number of characteristic endgroups per molecule and N is the number of moles of specific reagent equivalent to a known weight w of the sample. This is a fundamental method and does not require calibration against other methods.

In addition to the general requirements for endgroup analysis, spectroscopic techniques impose further conditions: (a) The compound being studied must contain a characteristic chromophore for which the molecular extinction coefficient is known. (The molecular extinction coefficients of the cellulose derivatives studied were determined from the corresponding derivatives of D-cellobiose.) (b) The characteristic chromophore should have an absorption maximum at a wavelength at which the remainder of the molecule does not absorb.

The purpose of the present study has been to investigate the potential of solvent Cadoxen as a spectroscopic solvent for cellulose and cellulose

derivatives, with particular reference to the possibility of estimating molecular weight by spectroscopic endgroup analysis.

EXPERIMENTAL

Materials

The reagents used were of Laboratory Standard grade and were used as received from the manufacturers.

Cotton cellulose (D.P. 3430) was used for the preparation of the hydrocellulose samples. The cotton was given a preliminary scour in 2% sodium hydroxide for 6 hr. This was followed by a 5-min sour in 0.5% sulfuric acid and a 5-min treatment with 0.25% sodium bicarbonate solution. Thorough rinsing was carried out after this treatment. Finally the cotton was extracted with an azeotropic mixture of absolute ethanol and chloroform.[9]

Ultraviolet and Visible Spectra

Ultraviolet and visible spectra were recorded with the Unicam SP 800 and SP 500 spectrophotometers, using matched 5-mm and 10-mm stoppered silica cells throughout.

Preparation and Analysis of Cadoxen

The Cadoxen was prepared by the procedure proposed by Donetzhuber,[10] which resulted in a Cadoxen solution containing 28% ethylenediamine and 5.3% cadmium and was 0.35*M* with respect to sodium hydroxide.

The cadmium content of the solvent was determined by complexometric titration with ethylenediaminetetracetic acid, following the procedure of Vink.[11] The ethylenediamine concentration was determined by titration with standard hydrochloric acid to the methyl red endpoint.

Preparation of Hydrocellulose Samples

A series of samples of purified cotton cellulose was treated with 1*M* sulfuric acid at 80°C for periods of 10 min to six days, thereby providing samples of suitable molecular weight range. After hydrolysis, the samples were rinsed with distilled water and then left overnight in a 0.25% sodium bicarbonate solution and finally rinsed well with distilled water.

Preparation of Cellulose Solutions

The cellulose solutions in Cadoxen were made up using a modification of the procedure of Henley.[4]

A finely chopped sample of cellulose was placed in a volumetric flask. The sample was wetted with a few drops of distilled water prior to dissolution. After a few minutes, the required volume of Cadoxen was added and the mixture was shaken until dissolution was complete. The solution was then made up to the mark by the gradual addition of distilled water.

The moisture content was determined as recommended by Lang and Mason,[12] using a separate portion of the sample, and the weight of the sample was corrected accordingly.

Determination of Molecular Weight of Hydrocellulose Samples

The molecular weights of the hydrocellulose samples were determined viscometrically as this method gives the most reproducible results.[13]

The flow times for the cellulose solutions were determined using a suspended-level dilution viscometer, fitted with a sintered glass filter disc of porosity 2. The viscometer was supplied by Polymer Consultants Limited, who state that the error arising from neglect of the kinetic energy correction is small for water, becoming vanishingly small for more viscous solvents such as Cadoxen.[14] As a result, no kinetic energy correction was made.

The limiting viscosity numbers $[\eta]$ were calculated using the method of least squares, and the limiting viscosity number–molecular weight relationships used were those proposed by Marx-Figini and Schulz,[15] namely:

$$\text{D.P.} = 274\ [\eta] \text{ for D.P. below 1000 and}$$

$$\log (\text{D.P.}) = 1.32 \log [\eta] + 2.25 \text{ for D.P. above 1000}$$

where $[\eta]$ is measured in dl/g. The results are shown in Table I.

TABLE I
Degree of Polymerization and Extinction Coefficients for Hydrocellulose Samples

Sample	$\overline{\text{D.P.}}_w$	ϵ at 239 nm
1	3432	17,790
2	1036	8,730
3	718	8,380
4	625	9,320
5	447	6,950
6	258	3,640
7	241	3,210

D-Cellobiose Phenylosazone

This was prepared according to the directions of Richtymer,[16] mp 207°C (lit.[16] 208°–209°C).

Hydrocellulose Phenylosazone

This was prepared according to the directions of Staud and Grey.[17]

RESULTS AND DISCUSSION

Hydrocellulose

Each hydrocellulose molecule contains one reducing endgroup, and if this endgroup has a characteristic absorption which differs from the absorption

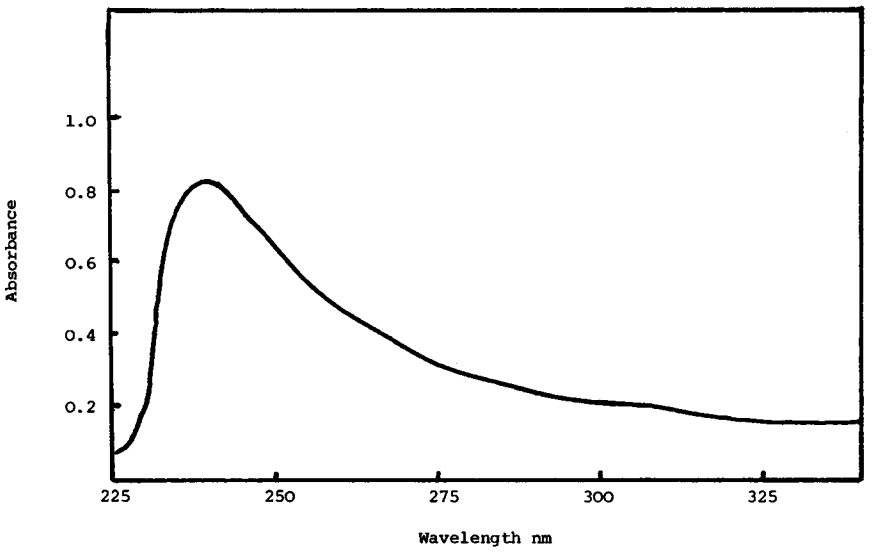

Fig. 1. Absorption spectrum of hydrocellulose in Cadoxen solution.

of the rest of the molecule, then it should in theory be possible to determine the molecular weight of hydrocellulose by spectroscopic endgroup analysis. Preliminary work has shown that this technique cannot be applied to hydrocellulose containing unmodified carbonyl endgroups.

Although the spectra of solutions of hydrocellulose in Cadoxen do show well-defined maxima at 239 nm (Fig. 1) and do obey Beer's law, there does not seem to be a simple relationship between the degree of polymerization and absorbance. Table I shows the extinction coefficients and degree of polymerization of six samples of hydrocellulose. The ultraviolet absorption by solutions of hydrocellulose in Cadoxen cannot be due solely to the reducing endgroup since, if this was the case, the molar extinction coefficient ϵ of each of the samples would be the same. Also, although the absorption spectra of D-glucose and D-cellobiose (Fig. 2) both show a maximum at 235–237 nm (D-glucose ϵ_{max} 37; D-cellobiose ϵ_{max} 104), their extinction coefficients differ considerably and are much lower than those for the hydrocellulose samples.

Several derivatives of hydrocellulose were studied which had strongly absorbing endgroups present. These included derivatives with phenylhydrazine, *p*-nitrophenylhydrazine, semicarbazide, thiosemicarbazide, hydroxylamine, and ethylacetoacetate. Of these, only the phenylosazone compounds were suitable.

Hydrocellulose Phenylosazone

Cellulose reacts with phenylhydrazine to give an osazone whose structure is similar to that of glucosazone and the osazones of oligosaccharides.[19]

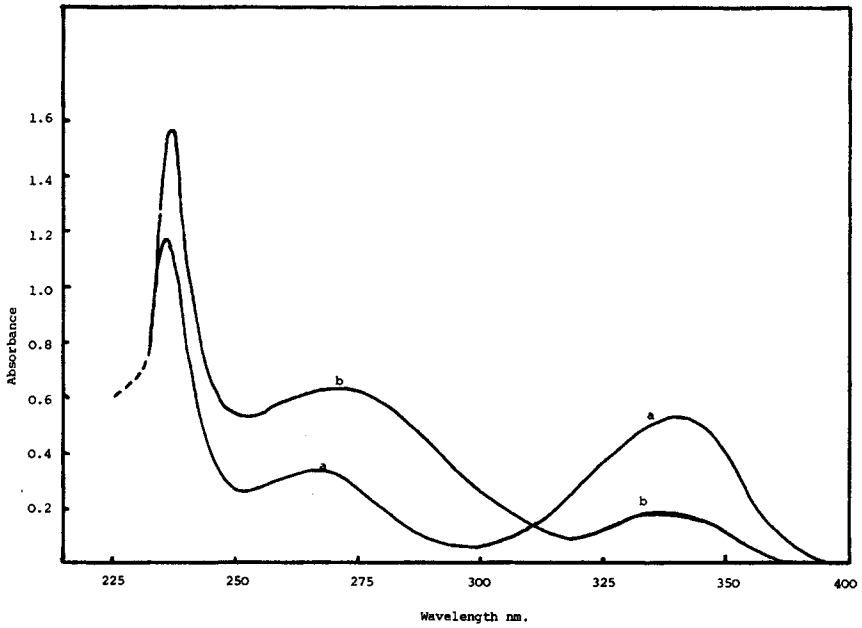

Fig. 2. Absorption spectra of (a) glucose and (b) cellobiose in Cadoxen solution.

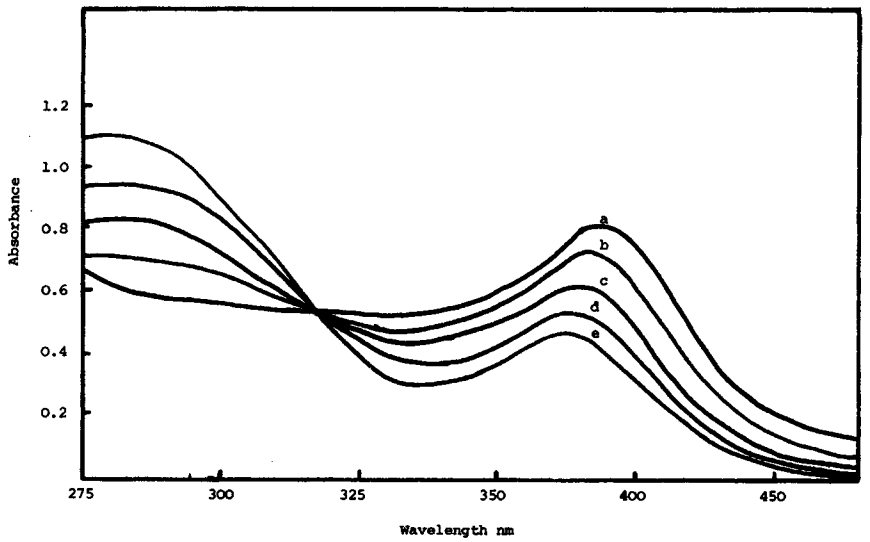

Fig. 3. Absorption spectrum of hydrocellulose phenylosazone in Cadoxen solution: (a) freshly prepared; (b), (c), (d), and (e) after 10, 20, 30, and 40 min, respectively.

The molar extinction coefficients of oligosaccharide phenylosazones are equal and independent of molecular weight[20] and satisfy the basic requirements of spectroscopic endgroup analysis. In addition, the absorption maxima of phenylhydrazine are well away from those of phenylosazones,

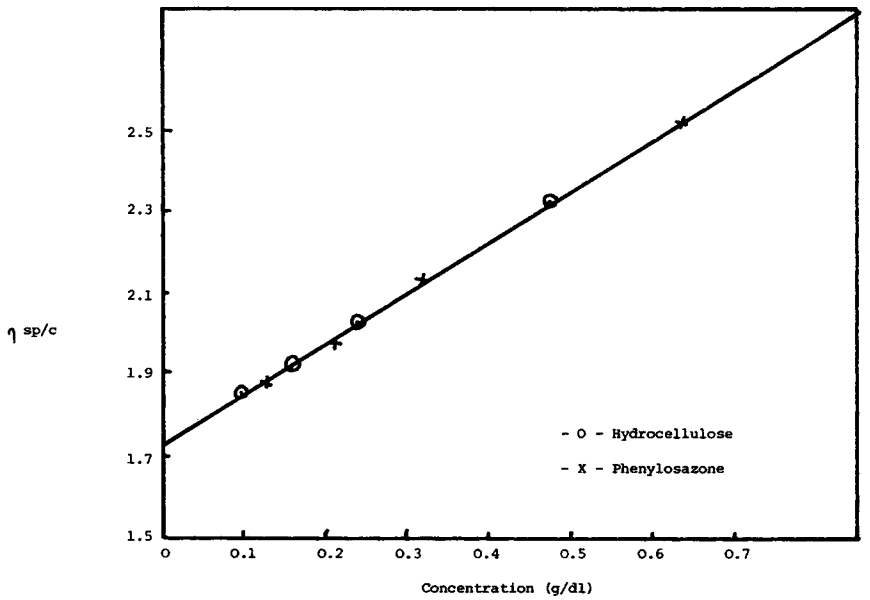

Fig. 4. Limiting viscosity number plot for hydrocellulose and hydrocellulose phenylosazone.

and so any phenylhydrazine bound by carboxyl groups will not interfere with the phenylosazone absorption spectrum.

A sample of hydrocellulose was treated with phenylhydrazine as described by Staud and Grey,[17] and after purifying by extraction with ethanol it was dissolved in 50% Cadoxen solution and its absorption spectrum determined. This shows two well-defined maxima at 277 nm and 390 nm, which, however, changed with time (Fig. 3). This change occurred in the presence of oxygen, and preliminary studies indicate that it is probably due to oxygenation followed by cleavage of phenylhydrazine residue. This effect is being investigated further. When the Cadoxen solvent is deoxygenated with nitrogen and the osazone solution prepared with the rigorous exclusion of oxygen, there is no observable change in spectrum during 1 hr, and this allows accurate measurements at 390 nm to be made.

To establish whether or not degradation occurs during reaction of the hydrocellulose with phenylhydrazine, the molecular weights of the hydrocelluloses were determined viscometrically and compared with the molecular weight of the phenylosazone. The results shown in Figure 4 indicate that no appreciable degradation occurs during the preparation of the derivative.

A series of phenylosazones was prepared using hydrocellulose samples of different D.P. Solutions of these were made in 50% Cadoxen and their absorption spectra determined. The results are shown in Table II. The average value of ϵ calculated from $\bar{M}_w$ is 14,390, which is close to that of cellobiose phenylosazone (ϵ = 14,490). However, for ϵ of the hydrocellulose phenylosazone to be compared with ϵ of cellobiose phenylosazone,

TABLE II
Molecular Weight, Degree of Polymerization, and Extinction Coefficients for Hydrocellulose Phenylosazones

Sample	$\overline{MW}_w$	$\overline{D.P.}_w$	ϵ at 390 nm
1	55.60×10^4	3,432	14,360
2	16.78 "	1,036	14,370
3	14.50 "	895	14,420
4	11.64 "	718	14,390
5	10.13 "	625	14,340
6	7.24 "	447	14,360
7	4.18 "	258	14,410
8	3.91 "	241	14,460

$\overline{M}_n$ should be used. The closeness of these two values is probably coincidence and may be explained by the fact that the reaction of the cellulose with phenylhydrazine does not go to completion. Treatment of the hydrocellulose samples for times greater than that recommended by Staud and Grey did not increase the phenylosazone content, and it was concluded that the maximum degree of reaction had been achieved.

The molar extinction coefficients of hydrocellulose phenylosazone and cellobiose phenylosazone varied slightly with different batches of Cadoxen, and so one batch of solvent must be used for each series of measurements. (The variation is probably due to small difference in cadmium content of the Cadoxen solvent.) However, as it is necessary to determine the molar extinction coefficient of a phenylosazone of known molecular weight, the variation from one batch of solvent to another does not complicate the endgroup analysis method.

The technique gives a useful spectroscopic method for the determination of the degree of polymerization of hydrocellulose.

We thank the Ministry of Education for Northern Ireland for a postgraduate studentship to R.C.

References

1. G. Jayme and F. Lang, in *Methods in Carbohydrate Chemistry*, Vol. III, R. L. Whistler, Ed., Academic Press, New York, 1963, Chap. 14.
2. H. F. Launer, and W. K. Wilson, *Anal. Chem.*, **22**, 455 (1950).
3. G. Jayme and K. Neuschäffer, *Naturwissenschaften*, **44**, 62 (1957).
4. D. Henley, *Svensk Papperstidn.*, **63**, 143 (1960).
5. A. Donetzhuber, *Svensk Papperstidn.*, **64**, 898 (1961).
6. A. Donetzhuber, *Svensk Papperstidn.*, **67**, 415 (1964).
7. E. Sjöström and B. Enström, *Svensk Papperstidn.*, **69**, 469 (1966).
8. S. R. Rafikov, S. A. Pavlova, and I. I. Tverdokhlebova, *Determination of Molecular Weights and Polydispersity of High Polymers*, Israel Program for Scientific Translations, Jerusalem, 1964, Chap. 8.
9. J. Wade and H. Finnemore, *J. Chem. Soc.*, **85**, 938 (1904).
10. A. Donetzhuber, *Svensk Papperstidn.*, **63**, 447 (1960).
11. H. Vink, *Makromol. Chem.*, **76**, 66 (1964).
12. A. R. G. Lang and S. G. Mason, *Can. J. Chem.*, **37**, 1829 (1959).

13. C. T. Greenwood, in *Physical Methods in Organic Chemistry*, J. C. P. Schwarz., Ed., Oliver and Boyd, Edinburgh and London, 1964, Chap. 7.
14. Polymer Consultants Limited, Technical Bulletin No. 4-V, London, 1962.
15. M. Marx-Figini and G. V. Schulz, *Makromol. Chem.*, **54**, 102 (1962).
16. N. K. Richtmyer, in *Methods in Carbohydrate Chemistry*, Vol. II, R. L. Whistler and M. L. Wolfrom, Eds., Academic Press, New York, 1963, p. 127.
17. C. J. Staud and H. le B. Grey, *Ind. Eng. Chem.* (Anal.), **1**, 80 (1929).
18. K. Christofferson and O. Samuelson, *Svensk Papperstidn.*, **65**, 517 (1962).
19. G. A. F. Roberts, Ph.D. Thesis, Queen's University, Belfast, 1966.
20. V. C. Barry, J. E. McCormick, and P. W. D. Mitchell, *J. Chem. Soc.*, 222 (1955).

Received May 1, 1972
Revised June 23, 1972