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AN INFRARED STUDY OF VARIOUS NITRATED POLYSACCHARIDES AND THEIR STRUCTURAL CHARACTERISATION

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ABSTRACT

Infrared absorption spectra for a number of polysaccharides and their nitrated derivatives have been obtained. The frequency range 730–960 cm^{-1} is useful for identification of the polysaccharides, and the region 900–1350 cm^{-1} is more suitable for distinguishing the nitrated materials. The strong intensity of the nitrate bands limits the interpretation of spectra below 960 cm^{-1}, but above this frequency the absorption bands of nitrated polysaccharides are generally sharper and more clearly defined than the corresponding bands of the parent polysaccharides. Data on the C–O–C bridge, C–C ring, C–O, and C–OH frequencies and on the C–H deformation and stretching frequencies have been obtained. The use of i.r. spectroscopy for the quantitative determination of nitrate groups in nitrated polysaccharides is discussed.

INTRODUCTION

I.r. spectroscopy is of value in the identification and analysis of compounds having similar molecular structure. Although it is difficult to assign specific group-functions to all bands in the spectrum of a complex molecule, much information regarding molecular structure can be obtained from absorption bands in the regions 4000–1350 and 900–650 cm^{-1}. The intervening zone, 1350–900 cm^{-1}, the so-called "finger print" region, frequently comprises a large number of bands whose origin is not easily determined[1], but which is of value in distinguishing closely related products such as anomeric forms of sugars and polymeric materials.

We now report on the i.r. spectra of a number of nitrated polysaccharides.

MATERIALS AND METHODS

The polysaccharides used in this study were commercial samples: three different types of pure dextrans (Koch–Light Laboratories), namely, food (mol. wt. $5\text{–}40 \times 10^6$), industrial (mol. wt. $2\text{–}3 \times 10^5$), and clinical grades (mol. wt. $6\text{–}9 \times 10^4$), with percentages of α-(1→6)-linked "anhydro-D-glucopyranose" units 94, 93, and 96.3%, respectively (*cf.* ref. 2); amylopectin (Koch–Light) and amylose (British Drug Houses); potato starch (BDH); cellulose (purified, bleached cotton-linters; 99.6% α-cellulose); α- and β-Schardinger dextrins (Koch–Light).

The polysaccharides were nitrated with four different reagents; the percentage of nitrogen in the products is given in brackets. (*a*) Fuming nitric acid (polysaccharide–acids ratio, 1:50 by weight) as described previously[3] (N, 13.3–13.4%). This nitrating reagent is not efficient for nitration of cellulose because of the hardening and gelatinization of the fibres[4]. (*b*) A mixture containing 50% of nitric acid and 25% each of acetic acid and acetic anhydride; this mixture minimizes depolymerization of polysaccharides and effects almost complete esterification[5] (N, 13.86–13.92%). (*c*) An anhydrous mixture of mixed acids, as described by Davis[6] for preparing starch nitrate, composed of 38.31% of nitric acid and 61.69% of sulphuric acid (material–acids ratio, 1:50 by weight) (N, 13.2–13.3%). (*d*) A mixed acid used industrially for preparing nitrocelluloses with a nitrogen content of 12.2% (N, 12.1–12.2%).

The crude nitrates were boiled for 3 h, and stabilisation was brought about by boiling for 72 h with distilled water followed by boiling with dilute, aqueous sodium carbonate for 48 h.

The i.r. measurements were performed on a high resolution Perkin–Elmer model 457 double-beam optical-null spectrometer scanning from 4000–250 cm^{-1} (2.5–40 μm). Discs were conventionally prepared from 1.5 mg of sample (previously dried *in vacuo* at 40° for 36 h) and 150 mg of potassium bromide; all spectra were obtained under quantitative conditions. In some cases, and particularly with fibrous samples that were difficult to grind, the sample discs tended to fracture when being removed from the die. To overcome this, it was necessary to use paper strengtheners, thereby adding to the sample thickness. It was noted that, where the thickness exceeded 43×10^{-2} mm, the disc lacked transparency with subsequent increase in background absorption.

RESULTS AND DISCUSSION

The spectra of the parent polysaccharides shown in Fig. 1 confirmed that the region 730–960 cm^{-1} is useful for the characterisation of D-glucose polymers[7,8]. The absorption peaks at 844 ± 8 and 891 ± 7 cm^{-1} allow the classification of the principal glycosidic linkages in a polyglucosan as α or β, respectively. In addition, from a consideration of the bands at 917 ± 13 and 766 ± 10 cm^{-1}, it is possible to obtain information about linkage positions. For example, whereas in the dextran spectrum the two absorptions at 917 ± 2 and 768 ± 1 cm^{-1} are typical of the α-(1$\rightarrow$6)-D-glycosidic linkages, in the starch-class spectra the absorptions at 930 ± 4 and 758 ± 2 cm^{-1} are typical of the α-(1$\rightarrow$4)-D-glycosidic linkages[7].

The spectra of the nitrated polysaccharides varied in quality. The stabilized products gave stronger absorptions and a higher degree of resolution. Purity of the polysaccharide sample markedly affected the quality of the spectra of the nitrated materials (*cf.* curves 3a and 3b in Fig. 1). Comparisons of spectra of unstabilized, polymeric carbohydrate nitrates may give misleading data and only the stabilized forms should be used. The stabilised esters obtained with the nitric acid–acetic

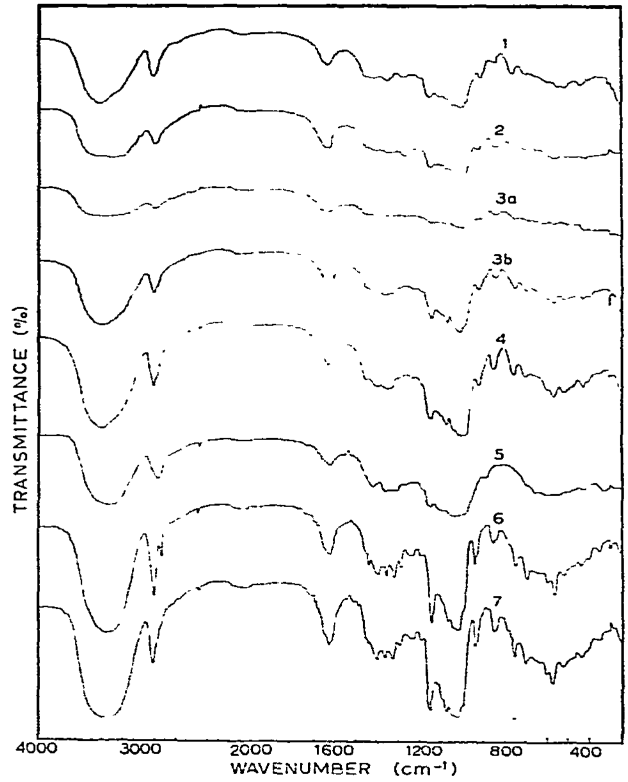

Fig. 1. Infrared spectra of un-nitrated polysaccharides. 1, clinical dextran; 2, amylopectin; 3a, amylose (Koch–Light); 3b, amylose (B.D.H); 4, starch; 5, cellulose; 6, α-Schardinger Dextrin; 7, β-Schardinger Dextrin.

acid–acetic anhydride mixture gave the best spectra, and Fig. 2 contains several examples.

From the spectra in Fig. 2 (*cf.* ref. 9), it is evident that the asymmetric and symmetric NO_2 frequencies are split and occur at ~1660 and ~1280 cm^{-1}, both bands being of strong intensity. Other absorptions attributable to the nitrate functional groups are at 840–850, 744, and 700 cm^{-1}. The spectra of nitrated polysaccharides (unstabilized or stabilized samples) prepared with mixed acids showed no extraneous absorptions compared to those prepared in the absence of sulphuric acid. Thus, there is no evidence to support the presence of absorptions due to sulphate ions[10]. The total sulphate contents[11] of unstabilized materials were 0.4–0.6% and 0.02% in the stabilized samples.

Comparisons of spectra of the polysaccharides and their nitrated derivatives showed that most of the absorption bands of nitrated materials in the region above 960 cm^{-1} are, in general, sharper and more clearly defined, whereas the region below 960 cm^{-1} (and especially that at 730–960 cm^{-1}, which is useful for characterising the

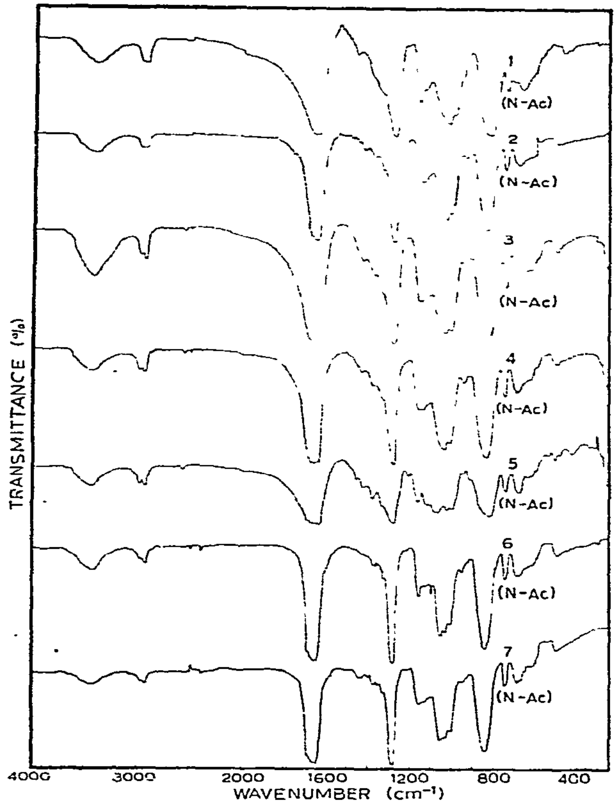

Fig. 2. Infrared spectra of nitrated polysaccharides. 1, dextran nitrate; 2, amylopectin (B.D.H.) nitrate; 3, amylose (B.D.H.) nitrate; 4, starch nitrate; 5, cellulose nitrate; 6, α-Schardinger Dextrin nitrate; 7, β-Schardinger Dextrin nitrate.

parent polysaccharides) is complicated by the presence of the strong nitrate bands which mask the weak to medium ring-CH vibrations.

The most important observations can be summarized as follows. On nitration, the intensity of the hydroxyl-stretching band (3200–3450 cm^{-1}) decreases and is shifted to a higher frequency by 30–40 cm^{-1}, probably by weakening of the hydrogen bonding[8].

In the C–H stretching region (2800–3000 cm^{-1}), a number of bands near 2850, 2920, and 2970 cm^{-1} were present in the spectra of most of the nitrated polysaccharides (Fig. 2). Thus, the C–H stretching band in the spectra of the parent polysaccharides (*e.g.* 2870 cm^{-1} for cellulose[12], and 2930 cm^{-1} for the other polysaccharides) indicates a number of partially overlapping bands that cannot all be assigned exclusively on a group-frequency basis, as has been previously noted[13].

The C–H deformation bands (1300–1480 cm^{-1}, see Fig. 2) are sharper than the corresponding bands of the parent polysaccharides (Fig. 1). In the spectrum of nitrated cellulose, the absorption at ~1380 cm^{-1} is considerably more intense than that of all the other, α-D-linked nitrated polysaccharides in which this particular vibration occurs as a shoulder. Thus, the band near 1380 cm^{-1} may be assigned to the C-1–H deformation vibration. The spectrum of cellulose nitrate exhibited a band at ~1460 cm^{-1} which was not present in the spectrum of the parent polymer and the assignment of which is unknown.

As expected, bands in the region (1000–1150 cm^{-1}) corresponding to the C–O–C bridge, C–C ring, C–O, and C–OH frequencies are broad. The spectra of the polysaccharides and the nitrated materials show medium to strong absorption at 1150–1160 cm^{-1} which is assigned to a ring frequency, probably to a bridge C–O–C stretching, but there is a possibility of the vibration being associated with the C–OH grouping (*cf.* ref. 13). From the nitration studies, it is evident that this absorption is not associated with the C–OH group since there is no frequency variation on nitration. All the parent polysaccharides showed one band of medium intensity at 1120 cm^{-1}, but it was not possible to establish whether this band contained two overlapping absorptions. From the spectra of the nitrated polysaccharides, however, two weak absorptions were identified at ~1110 and ~1130 cm^{-1}. The former band was assigned to an association band involving C–O stretch and the latter to a ring vibration, probably to an antisymmetrical C–O–C stretch, which would give rise to a parallel band[13].

The strong intensity of the nitrate absorption-bands severely limits interpretation of the region 730–960 cm^{-1}. However, the range 900–1350 cm^{-1} may be regarded as the "finger print" region for polysaccharide nitrates. In the nitrated dextrans, an absorption at 950 cm^{-1} appears as a shoulder which is not shown by nitrated amylopectin, and this band may be assigned to a ring vibration characteristic of the dextran α-(1→6)-D-linkage. In contrast, a weak absorption at 930–940 cm^{-1} is shown by nitrated amylopectin, at 940–950 cm^{-1} by nitrated amylose and nitrated starch, and at 960 cm^{-1} by the nitrated α- and β-Schardinger dextrins, all of which contain α-(1→4)-D-glycosidic linkages. The absorption near 910 cm^{-1} is characteristic of nitrated cellulose.

The region 980–1200 cm^{-1} is of limited value for distinguishing nitrated polysaccharides (Fig. 2). Thus, the dextrans, Schardinger dextrins, and cellulose nitrates can be distinguished from amylose, amylopectin, and starch nitrates but not from each other. With the exception of dextran nitrate, all the nitrated polysaccharides showed two absorptions near 1000 and 1040 cm^{-1}, whereas nitrated cellulose exhibited one more clearly defined band at 1070 cm^{-1}. The nitrated Schardinger dextrins had weak absorptions at 1055 cm^{-1} (Fig. 2). Nitrated dextran has absorptions at 980 and 1020 cm^{-1}, a shift of some 20 cm^{-1} compared to the absorptions of the other nitrated polysaccharides. Although assignments cannot yet be made, this region provides a convenient means for identifying the nitrated dextran molecule. Differentiation between the spectra for nitrated amylose and nitrated starch is impossible, but amylopectin, as shown above, can be characterized by the 930–940 cm^{-1} absorption.

The observed OH stretching vibrations varied inconsistently with the apparent degree of nitration (as determined by chemical analysis[14]), and hence the OH to CH ratio will not provide the necessary correlation for the quantitative determination of degree of nitration nor for the calculation of the nitrate extinction coefficient using the modified Beer's law equation[15]. The discrepancy observed in the OH to CH ratio appears to be due to the samples' becoming hydrated either during sample preparation or in the spectrometer prior to recording of spectra.

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