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Short communication

Determination of degree of substitution in succinic anhydride modified cellulose by headspace gas chromatography

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ABSTRACT

This paper reports on a headspace gas chromatographic method (HS-GC) for rapid determination of degree of substitution in succinic anhydride (SA) modified celluloses. The method is based on the reaction between the carboxyl groups in SA modified cellulose and bicarbonate solution in a closed headspace sample vial. The CO_2 released from the reaction was measured by HS-GC. The completion of the reaction was achieved within 25 min at 80 °C when a small sample size (<20 mg) was used. The relative standard deviation (RSD) measurement of precision was less than 4.1%, and the results were within 8.0% of those obtained with the traditional titration for determining the degree of substitution. The present method is simple, practical, automated, and suitable for use in anhydride modified cellulose research.

1. Introduction

Cellulose is a plant-based polymer mainly used to produce paperboard and paper. Modification of cellulose by reacting other molecules onto its structure gives the material greater flexibility for use in many advanced applications [1–4]. Esterification of cellulose with anhydrides is an important step in introducing functional groups, such as alkyl, acyl or carboxyl groups, onto cellulose [5,6] so that other desired molecules can be grafted on the structure in later steps. The degree of substitution (DS), i.e., the average number of the functional groups bonded in an anhydroglucose unit on the cellulose backbone resulting from substituting on its hydroxyl groups, is an important parameter in cellulose modification research and industrial applications. For example, the DS is closely related to the properties of the final products, such as thermoplasticity, water absorbency and performance in drug delivery [7–9].

Succinic anhydride (SA) is an effective carboxylating agent in cellulose acylation [10] and, therefore, is widely used in cellulose modifications. The DS of SA on cellulose can be calculated if the content of carboxyl groups on cellulose is available. Although there are many methods for quantifying the content of carboxyl groups on cellulose, some of the methods suffer from the technical difficulties that limit their application, e.g., incomplete solubility of the sample in a deuterated solvent used in nuclear magnetic resonance (NMR) analyses [11] or a water content interference in the Fourier transform infrared spectroscopic (FTIR) analyses [12]. Alkaline hydrolysis, coupled with acid back titration, gas chromatography (GC) and liquid chromatographic method (HPLC), is a method commonly used to quantify carboxyl groups on polysaccharide polymers, such as cellulose and starch [13–16]. Titration methods were also applied to determine the DS of the other acidic cellulose polymers such as cellulose phthalates [17,18] and cellulose acetates [19]. The major disadvantage in these methods is that they require a long time for alkaline hydrolysis and involve several procedures in the testing, which makes the methods less efficient.

Headspace gas chromatography (HS-GC) has been found to be a useful analytical technique to analyze volatile species in complex matrices, including solids [20,21]. A distinguishing advantage of HS-GC is that the headspace sample vial can be used as a mini-reactor, in which the non-volatile species can be chemically converted to corresponding volatile species that can be easily analyzed by GC, leading the direct or indirect determination of the analyte of interest [22]. Moreover, since HS-GC can perform an in situ and automatic measurement after the reaction for multiple samples, it makes the method much simpler and efficient. According to our previous studies [23,24], the carboxyl acids in pulp fibers react with bicarbonate to form carbon dioxide (CO_2), which can be easily measured by HS-GC equipped with a thermal conductivity detector. We believe that the carboxyl groups on SA modified cellulose (Cell-SA) can also be determined indirectly by the similar approach mentioned above.

In this paper we present a HS-GC method for the determination of carboxyl groups on Cell-SA, from which the DS of Cell-SA can be determined. The focus is on the effects of the major experimental conditions; e.g., the reaction temperature and time, on the HS-GC measurement.

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2. Experimental

2.1. Materials

Cotton cellulose, was purchased from Sinopharm Chemical Reagent Co. Ltd. Succinic anhydride (SA), N,N′-dimethylacetamide (DMAc), lithium chloride (LiCl), triethylamine (TEA) and 4-dimethylaminopyridine (DMAP) were of all analytical grade and purchased from Aladdin Reagent Co. Ltd.

2.2. Instruments and operations

All measurements were carried out using a headspace sampler (DANI HS 86.50, Italy) and gas chromatograph (Agilent HP-7890, Palo Alto, CA, USA) equipped with a thermal conductivity detector (TCD). Headspace sampler operating conditions were as follows: oven temperature = 60–80 °C; vial pressurization time = 0.2 min; sample-loop fill time = 0.2 min; loop equilibration time = 0.05 min; and loop fill time = 0.2 min. The GC was equipped with a model GS-Q capillary column with an i.d. of 0.53 mm and a length of 30 m from J&W Scientific, Folsom, CA. It was operated at 45 °C with nitrogen as the carrier gas at a flow rate of 3.1 mL/min, with a TCD set at 220 °C.

2.3. Procedures for sample preparation and HS-GC measurement

A roughly 15 mg sample of a Cell-SA sample (finely powdered), was accurately weighed and placed in a 20 mL headspace sample vial. After the addition of 4 mL of bicarbonate solution (0.02 mol/L), the vial was immediately sealed by a septum, shaken to obtain a good dispersion in the solution, and placed in the headspace sampler for automatic HS-GC testing.

2.4. Determination of DS by a reference titration method

A known weight of the sample was dissolved in 50 mL of NaOH (0.500 mol/L) and stirred for 60 min at 50 °C. The excess amount of NaOH was back titrated with a standard HCl solution (0.250 mol/L). The DS was calculated by the following equation [13–15]:

$$DS = \frac{162 \times n_{COOH}}{m - 100 \times n_{COOH}} \quad (1)$$

where $n_{COOH} = (V_{NaOH} \times C_{NaOH} - V_{HCl} \times C_{HCl})/2$, m, 162, and 100 are the molar number of carboxyl groups, the sample weight, the molar mass of anhydroglucose unit (AGU), and the net increase in the mass of an AGU for each succinic substituted, respectively.

3. Results and discussion

3.1. Reaction temperature and time for carboxyl neutralization

The present work is based on the reaction between the carboxyl groups in Cell-SA and bicarbonate, i.e.,

$$RCOO(CH_2)_2COOH + HCO_3^- \rightarrow RCOO(CH_2)_2COO^- + H_2O + CO_2 \quad (2)$$

in which R represents the cellulose backbone. If the above reaction is complete, the amount of carboxyl groups in Cell-SA can be indirectly quantified by measuring the CO_2 generated from the reaction. Since only air and CO_2 presented in the headspace of sample vial, the chromatogram is very simple. As observed, two well separated peaks (i.e., O_2 and CO_2), at the retention time of 2.370 and 3.091 min respectively, were shown in the chromatographic run.

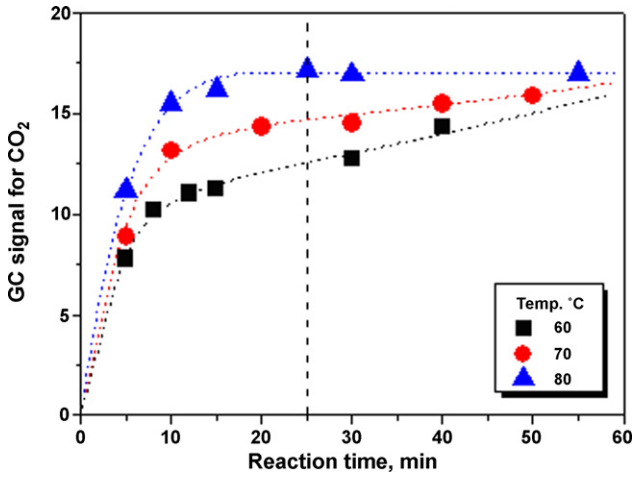

Fig. 1. Effect of reaction temperature and time on the neutralization of carboxyl groups in Cell-SA.

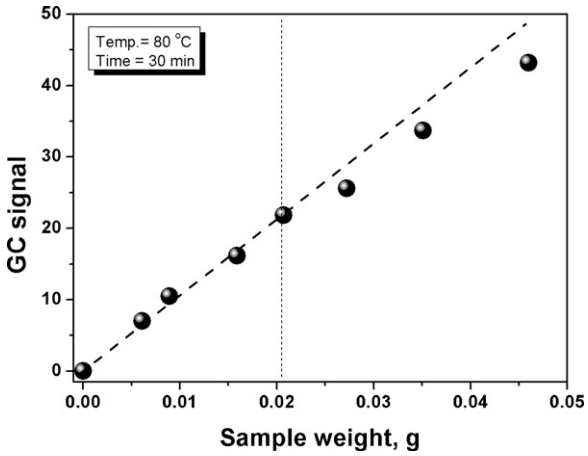

Fig. 2. Effect of sample size on carboxyl conversion in the neutralization reaction.

As shown in Fig. 1, both the temperature and time were important in determining the completion of the reaction at the given bicarbonate solution (4 mL of 0.02 mol/L). At a temperature of 80 °C, the reaction reached completion within 25 min or earlier. At a lower temperature, the reaction reached completion more slowly. According to our previous work for pulp fibers (the amount of carboxyl groups is less than 100 μmol/g [22,23]), the complete reaction can be achieved within 10 min at 60 °C. However, a longer reaction time even performed at 80 °C was required in the present work. We believe that due to higher amount of carboxyl acids in Cell-SA, the swelling time is important for the agglomerated sample in the reaction medium, which affects the completeness of the neutralization reaction between bicarbonate and the polymer's carboxyl acids.

3.2. Sample size

For complete carboxyl acid neutralization, it is essential to have an excess amount of bicarbonate in the reaction system. In the given bicarbonate solution (4 mL of 0.02 mol/L) used in this work, the absolute content of bicarbonate was 0.08 mmol. Thus, the molar amount of carboxyl groups in the Cell-SA samples should be lower than that of bicarbonate. Fig. 2 shows the GC responses to the CO_2 converted from carboxyl acid neutralization when different weights of the Cell-SA sample (carboxyl content = 980 μmol/g) were added at 80 °C for 30 min. It is observed that to a complete

Table 1
Reproducibility in HS-GC measurement.[a]

Sample ID	Replica No.			Mean value	RSD, %
	1	2	3		
1	40.0	37.1	37.6	38.2	4.05
2	19.4	20.2	20.3	20.0	2.47
3	35.4	34.3	33.2	34.3	3.21

[a] Sample size = 15 mg.

Table 2
Method comparison.

Sample ID	Carboxyl groups, μmol/g		Difference %
	HS-GC	Titration	
1	767	762	+0.65
2	1500	1490	+0.67
3	1590	1510	+5.16
4	2060	1910	+7.55

reaction the sample size should be no greater than 0.02 g; otherwise, the reaction might not be completed at the given conditions. For 0.02 g of the above Cell-SA sample, the absolute content of carboxyl is about 0.02 mmol, which is 4 times lower than that of bicarbonate (0.08 mmol). Thus, we conclude that the overdosed bicarbonate is necessary for a complete carboxyl conversion in Eq. (2) at 80 °C for 30 min. If the carboxyl content in the Cell-SA sample is higher, then the sample size should be smaller.

3.3. Method calibration

The method was calibrated by spiking different volumes (0–100 μL) of a standard HCl solution (0.500 mol/L) into a set of closed headspace sample vials containing 4 mL of bicarbonate solution. The headspace sampler automatically shakes the sample (to facilitate the neutralization reaction) and uses GC to measure the CO_2 gas released in the vials. Because there is a small amount of CO_2 in the headspace of the vials (from air and bicarbonate decomposition [23,24]), a blank solution (without a sample) was run to account for their effects.

The relationship between the GC responses (signals) and CO_2 released from the reaction shown in Fig. 2 can be written as

$$A = 0.0207(\pm 0.0310) + 961(\pm 12.6) \times C \quad (n = 6, r^2 = 0.9993) \quad (3)$$

where A is the net GC response to the CO_2 released from the reaction and C is concentration of CO_2 (in mmol).

The limit of quantitation (LOQ) for CO_2 is 0.344 μmol (equals to 3.72×10^{-3} for DS of SA in cellulose), calculated from the following equation [25].

$$LOQ = \frac{a + 10 \times |\Delta a|}{s} \quad (4)$$

where a, Δa and s represent the intercept, uncertainty of the intercept, and slope in Eq. (3), respectively.

3.4. Method evaluation

3.4.1. Precision

The precision of the present method was evaluated by triplicate analysis of three Cell-SA samples. The results showed that the relative standard deviations (RSD) of these measurements were less than 4.1% (see Table 1).

3.5. Validation

The present method was validated by a comparing the results of the HS-GC method with the results determined by the titration method [15]. As Table 2 shows, the differences for carboxyl quantification were within 8.0% of each other. It is noticed that the amounts of carboxyl measured by the present HS-GC method are always higher than those measured by the titration method. In order to check if the hydrolysis of the ester substituent from the cellulose back bone took place at the given reactions (30 min at 80 °C) conditions in the present method, an ion chromatographic (IC) measurement was performed for detecting the succinate in the filtrates (passing through a 0.2 μm membrane) of the sample solution after the neutralization. The results showed that the amounts of succinate acid in all these sample filtrates were very low, which are less than 0.5% of the carboxyl acids in the sample listed in Table 2. Thus, we can conclude that the positive interference in the present method is insignificant. Therefore, the lower results from the titration method we conducted (reaction at 50 °C for 60 min) was probably caused by both the incompletion of alkaline hydrolysis and the precipitation of the fatty acids during the titration performed at room temperature [16]. Therefore, the present method is more justifiable than the titration method to be used for the determination of the carboxyl groups in Cell-SA samples.

4. Conclusion

A HS-GC method for the determination of carboxyl content in Cell-SA has been developed. The present method has good measurement precision (RSD < 4.1%). The results of this method were within 8.0% of the results obtained with the standard titration method for DS determination. The present method is simple, practical, automated, and suitable for use in anhydride modified cellulose research.

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