Original paper

Interconversion Between d-glucuronic Acid and d-glucuronolactone in Subcritical Aqueous Ethanol

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\(\text{d-glucuronic acid (GlcA) and d-glucuronolactone (GlcL) were treated with subcritical aqueous ethanol in the range of 0\% to 80\% (w/w) at 180^\circ\text{C} in order to examine the effect of ethanol on the interconversion between GlcA and GlcL. When GlcA was treated at higher ethanol concentrations, less GlcA disappeared and more GlcL was formed compared to treatments at lower ethanol concentrations. For comparison, in the treatment of GlcL at higher ethanol concentrations, disappearance of GlcL was slower and less GlcA was formed. The degradation and interconversion of GlcA and GlcL were kinetically analyzed under the assumptions of first-order kinetics in order to evaluate the rate constants for each process. The rate constants were smaller at higher ethanol concentrations. This was particularly manifested in the significantly low rate constants observed at ethanol concentrations higher than 60\%. The treatments of GlcA and GlcL were also examined at 200^\circ\text{C}, and the effect of ethanol at 200^\circ\text{C} was similar to that at 180^\circ\text{C}.}

Keywords: glucuronic acid, glucuronolactone, subcritical water, ethanol

Introduction

Subcritical water, i.e., water that maintains its liquid state at elevated temperatures under pressurized conditions, has two unique features. One is a lower relative dielectric constant, which is almost the same as those of ambient methanol and acetone (Yang et al., 1998; Eckert et al., 2000). This low dielectric constant facilitates the use of subcritical water to extract hydrophobic substances from various resources (Hashimoto et al., 2004; Rodriguez-Meizoso et al., 2006). Another feature is the high ion product. Since this fact denotes a high concentration of hydrogen and hydroxyl ions, subcritical water can subsequently act as an acid or base catalyst (Kuhlmann et al., 1994; Lesutis et al., 1999; Kramer and Vogel, 2000). Based on these properties, extraction (Ravber et al., 2015), hydrolysis (Sasaki et al., 1998), and isomerization (Usuki et al., 2007) in subcritical water have been reported.

Processes using subcritical water should be environmentally friendly and have no adverse effects on the human body. Therefore, the use of subcritical water is promising for the environmentally conscious and effective utilization of biomass through its degradation (Miyazawa and Funazukuri, 2004; Liu and Wyman, 2005; Garna et al., 2006). Since biomass is hydrolyzed by subcritical water to its constituent pentoses, hexoses, and hexouronic acids without the need for additional acids or bases, hydrolysis by subcritical water does not require neutralization of acid or base reactants.

\(\text{d-glucuronic acid (GlcA) is a hexouronic acid obtained through}

the hydrolysis of hemicellulose, which is a major component of biomass. GlcA is converted to \(\alpha\)-glucuronolactone (GlcL) in subcritical water (Wang et al., 2010). We previously reported that the addition of ethanol to water significantly promotes the isomerization of mono- and disaccharides under subcritical conditions (Gao et al., 2014; 2015a; 2015b; 2015c; 2016). However, it remains unclear which reactions will occur during the treatment of GlcA in subcritical aqueous ethanol. In this context, the interconversion between GlcA and GlcL in subcritical aqueous ethanol is examined at relatively low temperatures (180°C or 200°C) and kinetically analyzed.

Materials and Methods

Materials \(\alpha\)-glucuronic acid (GlcA, purity >98%) was purchased from Wako Pure Chemical Industries (Osaka, Japan). \(\alpha\)-Glucurono-6,3-lactone (GlcL, purity >99%) was obtained from Sigma-Aldrich Japan (Tokyo).

Treatment of \(\alpha\)-glucuronic acid or \(\alpha\)-glucuronolactone in subcritical aqueous ethanol GlcA or GlcL was dissolved in distilled water and was then mixed with ethanol to produce the solution with a final GlcA or GlcL concentration of 0.5% (w/w). The ethanol concentration was varied in the range from 0 to 80% (w/w). The feed solution was sonically degassed before the subcritical treatment and was connected to a nitrogen gasbag to prevent re-dissolution of atmospheric oxygen. The feed solution was delivered into a coiled stainless steel (SUS 316) tubular reactor (0.8 mm I.D. \(\times\) 2.0 m length) immersed in SRX 310 silicone oil (Toray-Dow-Corning, Tokyo, Japan) with a residence time of 10 to 240 s by an L-7100 high-performance liquid chromatographic (HPLC) pump (Hitachi, Tokyo, Japan). The residence time was calculated based on the inner diameter and length of the stainless steel tube and the density of the water-ethanol mixture under subcritical conditions, in reference to an earlier study (Wang et al., 2010). The reaction was conducted at both 180°C and 200°C. The reactor effluent was directly introduced to a stainless steel tube (0.8 mm I.D. \(\times\) 1.0 m length) immersed in an ice-water bath (to terminate the reaction) and was collected in a sampling vessel. The pressure inside the tube was regulated at 10 MPa by a back-pressure regulator (high pressure adjustable BPR P-880; Upchurch, WA, USA). The effluent of a fixed volume (usually 0.20 mL) in a sampling vessel was evaporated under reduced pressure. The remainder was dissolved with distilled water of the same volume to prepare the sample for HPLC analysis.

HPLC analysis The concentrations of GlcA and GlcL in the effluent were determined using an HPLC consisting of a pump (L-7100, Hitachi, Tokyo, Japan), a COSMOSIL Hilic column (3.0 mm I.D. \(\times\) 150 mm, Nacalai Tesque, Kyoto, Japan), and a refractometer (L-3350, Hitachi). A mixture of 10 mmol/L CH₃COONH₄ and acetonitrile (50/50, v/v) was used as the eluent at a flow rate of 0.4 mL/min. The column temperature was maintained at 30°C in an L-7300 column oven (Hitachi).

Results and Discussion

Treatment of \(\alpha\)-glucuronic acid or \(\alpha\)-glucuronolactone in subcritical aqueous ethanol at 180°C The 0.5% (w/w) GlcA samples dissolved in 0% to 80% (w/w) aqueous ethanol solutions were heated at 180°C, and the disappearance of GlcA and the formation of GlcL were observed (Fig. 1). The disappearance of GlcA was decelerated at higher ethanol concentrations, while the effect of ethanol content on the formation of GlcL was complicated. The formation of GlcL was largely suppressed at 80% (w/w) ethanol. The formation of GlcL would proceed through acid-catalyzed lactonization. Because the pKₐ value of ethanol is higher than that of water (Wyatt, 2014), the ion product of aqueous ethanol and dissociation of GlcA would change. As a result, the concentration of hydrogen ion would change. This effect may remarkably appear at 80% (v/v) ethanol for the suppression of GlcL formation. For other concentrations, the formation rate of GlcL did not significantly depend on the ethanol concentration, although the disappearance of GlcL seemed to be faster at lower ethanol concentrations at long residence times. This fact would be related to the suppression of GlcL at higher ethanol concentrations as described later.

Figure 2 shows the disappearance of GlcL and the formation of GlcA when 0.5% (w/w) GlcL was treated at 180°C in aqueous solutions with various ethanol concentrations. The disappearance of GlcL and the formation of GlcA were decelerated and suppressed, respectively, at higher ethanol concentrations. Comparison of the results shown in Fig. 2 to those shown in Fig. 1 revealed that the disappearance of GlcL was much slower than that of GlcA, and the formation of GlcA was very small when GlcL was treated in aqueous ethanol. These results indicate that the interconversion between GlcA and GlcL occurs reversibly; however, the conversion of GlcL to GlcA is preferred.

Kinetic analysis Based on our previous study on the interconversion between GlcA and GlcL in subcritical water (Wang et al., 2010), the results shown in Figs. 1 and 2 were kinetically analyzed. All of the reactions shown in Fig. 3 were assumed to obey first-order kinetics. The formation rates of GlcA and GlcL are formulated by Eqs. (1) and (2), respectively.

\[
\frac{dC_A}{dt} = -(k_{AL} + k_{Ad})C_A + k_{LA}C_L \quad \cdots \text{Eq. 1}
\]

\[
\frac{dC_L}{dt} = k_{AL}C_A -(k_{LA} + k_{Ld})C_L \quad \cdots \text{Eq. 2}
\]

where \(C\) is the concentration, \(t\) is the residence time in the tubular reactor, and \(k\) is the first-order rate constant. The subscripts A and L represent GlcA and GlcL, respectively. The subscripts AL and LA indicate the conversion of GlcA to GlcL and the reverse conversion, respectively, and \(d\) represents degradation.

The rate constants, \(k_{AL}, k_{LA}, k_{Ad}\), and \(k_{Ld}\) at each ethanol concentration were estimated to minimize the sum of the residual squares between the experimental and calculated values of \(C_A\) and \(C_L\) for both the reactions in Figs. 1 and 2 using the Solver of
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Microsoft Excel® 2010. The calculated concentrations were obtained by solving Eqs. (1) and (2) simultaneously by the numerical Euler method where the increment in \( \tau \) was set to 1 s.

Figure 4 shows rate constants, \( k_{AL} \), \( k_{LA} \), \( k_{Ad} \) and \( k_{Ld} \), at various ethanol concentrations. The rate constants for the degradation of GlcA and GlcL, \( k_{Ad} \) and \( k_{Ld} \), were smaller at higher ethanol concentrations. In other words, the addition of ethanol into water suppressed the degradation of GlcA and GlcL. The rate constants for the interconversion between GlcA and GlcL, \( k_{AL} \) and \( k_{LA} \), were also smaller at higher ethanol concentrations. The effect of ethanol on the rate constants became remarkably small at ethanol concentrations higher than 60% (w/w). Many factors affected the rate constants. All the rate constants reflect the concentration of hydrogen ion. The concentration of hydrogen ion would vary with the change in the ethanol concentration. Therefore, one of the reasons for the change in the rate constants would be related to the change in the ethanol concentration of the solvent. However, it remains unclear whether the higher ethanol concentration or the lower water concentration predominantly affects the smaller rate constants at higher ethanol concentrations.

Treatment at 200°C GlcA, which was dissolved at 0.5% (w/w) in water, 40% (w/w) ethanol, or 80% (w/w) ethanol, was also treated at 200°C (Fig. 5). The disappearance of GlcA and the formation of GlcL were both slower at higher ethanol concentrations. This trend was similar to that at 180°C, although the degradation of GlcL was faster at 200°C than at 180°C. The 0.5% (w/w) GlcL dissolved in water was also treated at 200°C (Fig. 4). The formation of GlcA from GlcL was small at 200°C and was similar to that at 180°C.
References


