Note

Solubility of D-Galactose, D-Talose, and D-Tagatose in Aqueous Ethanol at Low Temperature

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Received June 14, 2015 ; Accepted August 20, 2015

The solubility of D-galactose, D-tagatose, and D-talose in aqueous ethanol (20 – 80 wt%) was measured in the temperature range of –30°C to 20°C. The solubility of each sugar decreased upon increasing the ethanol concentration and decreasing the temperature. The solubility of D-talose and D-tagatose was higher than that of D-galactose under all conditions. The temperature dependence of the solubility of each hexose can be expressed by the van't Hoff equation. The dissolution enthalpy of each hexose decreased upon increasing the ethanol concentration. The dissolution enthalpy of D-talose was higher than that of D-galactose and D-tagatose.

Keywords: D-galactose, D-talose, D-tagatose, solubility, aqueous ethanol

Introduction

D-Tagatose and D-talose, both of which are rare sugars, have many important uses such as food additives to improve health and extend life span (Levin *et al.*, 1995; Lu *et al.*, 2008) and raw materials to synthesize value-added products (Baer, 1962; Yu and Chung, 1968). As a result, many methods such as alkali-, metal-catalyzed, and enzymatic isomerization have been developed to produce D-tagatose and D-talose (Khadem *et al.*, 1989; Yamauchi *et al.*, 1990; Lim *et al.*, 2007; Beerens *et al.*, 2012). Among these methods, the alkali-catalyzed isomerization of D-galactose is promising for use in the food industry for reasons of food safety, pretreatment, and postpurification, although many byproducts are produced through side reactions.

We previously reported that high yields of D-tagatose can be produced by treating D-galactose with subcritical aqueous ethanol, along with the production of a small amount of D-talose (Gao *et al.*, 2015). This method is also considered promising for the production of D-tagatose from D-galactose because of its high productivity and short reaction time. This method produces unreacted substrates, desired products, and some byproducts in the reaction effluent.

Therefore, to reduce the load on the separation process, decreasing the temperature or increasing the ethanol concentration for precipitating desirable or undesirable components before chromatographic separation is considered a possible solution as the first step in the purification process.

In this study, we measured the solubility of D-galactose, D-tagatose, and D-talose in aqueous ethanol in the temperature range of –30°C to 20°C.

Materials and Methods

Materials Ethanol, D-galactose, D-tagatose, and D-talose were purchased from Wako Pure Chemical Industries (Osaka, Japan). In this study, the solubility of only D-enantiomers was investigated, and therefore, the prefix, D-, of all hexoses is omitted hereafter.

Solubility measurement The solubility of galactose, tagatose, and talose in aqueous ethanol (20 – 80 wt%) was measured in the temperature range of –30°C to 20°C. The temperature was controlled using an FP89-HL ultralow refrigerated-heating circulator (Julabo Japan, Osaka, Japan). A solution of galactose, tagatose, or talose was prepared in a vial by adding an excess

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Fig. 1. Temperature dependence of solubility of (a) galactose, (b) talose, and (c) tagatose in (◇) 20 wt%, (□) 40%, (△) 60%, and (○) 80% ethanol.

Table 1. Dissolution enthalpies of galactose, talose, and tagatose, ΔH_i , in aqueous ethanol.

Ethanol concentration [wt%]	ΔH_i [kJ/mol]		
	Galactose	Talose	Tagatose
20	10	16	8.6
40	3.9	17	6.7
60	3.1	11	3.9
80	3.0	6.9	1.3

amount of hexose to aqueous ethanol. The solution was kept at the measuring temperature for 24 h to ensure equilibrium solubility.

The supernatant of the hexose solution was diluted to a detectable concentration for HPLC quantification with aqueous ethanol. The HPLC system consisted of an LC-10AD VP HPLC pump (Shimadzu, Kyoto, Japan), RI-101 refractometer (Showa Denko, Tokyo, Japan), and a Cosmosil Sugar-D column (4.6 mm I.D. $\times$ 250 mm length; Nacalai Tesque, Kyoto, Japan). The column was maintained at 30°C in a CTO-10A VP column oven (Shimadzu). A water-acetonitrile (20:80, v/v) mixture was used as the eluent at a flow rate of 1.0 mL/min.

Results and Discussion

Figures 1(a), (b), and (c) show the solubility of galactose, talose, and tagatose in 20–80 wt% aqueous ethanol at various temperatures according to the van't Hoff equation:

$$\frac{d \ln S_i}{d(1/T)} = -\frac{\Delta H_i}{R} \quad \text{.....Eq. 1}$$

where S_i is the solubility of hexose i ; ΔH_i , the dissolution enthalpy of hexose i ; R , the gas constant; T , the absolute temperature; and subscript i , galactose, talose, or tagatose. The solubility of all hexoses decreased upon increasing the ethanol concentration and decreasing the temperature. For example, at 20°C, the solubility of galactose, talose, and tagatose decreased from 0.093 to 0.003, 0.502 to 0.057, 0.527 to 0.048 mol/kg-solvent, respectively, upon changing the solvent from 20 wt% aqueous ethanol to 80 wt% aqueous ethanol. The solubility of galactose and tagatose in 40 wt%

aqueous ethanol both decreased by ~40% upon decreasing the temperature from 20°C to –30°C, and by ~66% from 20°C to –20°C for talose. Further, the hexose solubility was more sensitive to temperature change at lower ethanol concentration (Alves *et al.*, 2007; Flood *et al.*, 1996), indicating that the solvent with low ethanol concentration was favorable for obtaining a solution with high tagatose content by hexose crystallization, which would be similar to the separation of fructose and glucose by freezing crystallization (Silva *et al.*, 2010). The solubility of talose and tagatose was much higher than that of galactose under all measured conditions. Hexoses exist as equilibrium mixtures of pyranose and furanose with α - or β -configurations. Therefore, the temperature dependence of the saccharide solubility cannot be simply explained. However, it was reported that the solubility of mannose and fructose in aqueous ethanol was higher than that of glucose at the same temperature and ethanol concentration (Hudson and Yanovsky, 1917). Because talose, tagatose, and galactose were the C-4 epimers of mannose, fructose, and glucose, respectively, this tendency may be also applicable to the difference in solubility of talose, tagatose, and galactose. The difference in solubility increased with increasing ethanol concentration. In particular, the solubility of talose was the most sensitive to the temperature change compared with that of galactose and tagatose.

The dissolution enthalpy, ΔH_i , of each hexose was evaluated from the slope of each line drawn in Fig. 1 according to Eq. (1). The estimated ΔH values are listed in Table 1. The dissolution enthalpy of any hexose decreased upon increasing the ethanol

concentration, and the dissolution enthalpy of talose was larger than that of galactose and tagatose.

These results indicate that talose and tagatose can be separated from galactose by adding ethanol, decreasing the temperature of the reaction mixture, or by the combined use of these two means.

Acknowledgements One of the authors (D.-M. G.) gratefully acknowledges a Monbukagakusho Scholarship from the Japanese government and the Tojuro Iijima Foundation for Food Science and Technology. This study was also partly supported by JSPS KAKENHI (No. 26870296; T. K.).

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