

## Measurement of Trehalose Dihydrate by Microscope Heating Method

### Introduction

Recently, trehalose (α-D-glucopyranosyl-α-D-glucopyranoside), one of disaccharides, has attracted attention as a sugar which protect biologic molecule under the dry condition. Some insects and plants become asphyxiated temporarily by drying, which could resurge by obtaining water. It is considered that trehalose is deeply related to this resurgence. Moreover, trehalose has been widely applied as additives for moisturizer in cosmetics and dry foods. However, the mechanism of bio-protecting functions by trehalose has not yet been known. To study the mechanism of bio-protecting functions, bound water in trehalose was measured using heated FTIR microscope.

### Systems and Method

Heating system (Mettler FP-81) was set on the XY stage of MFT-2000. Trehalose dihydrate was compressed between 2 KBr plates (7 × 7 × 1 mm), and tablet was formed using 10 mm KBr pellet die with 10 t press. The tablet was set in the heating system, and measured by microscope transmittance method with temperature ramping rate of 1°C/min from 60°C to 80°C. Neotrehalose hydrate (α-D-glucopyranosyl-β-D-glucopyranoside) was used as reference.

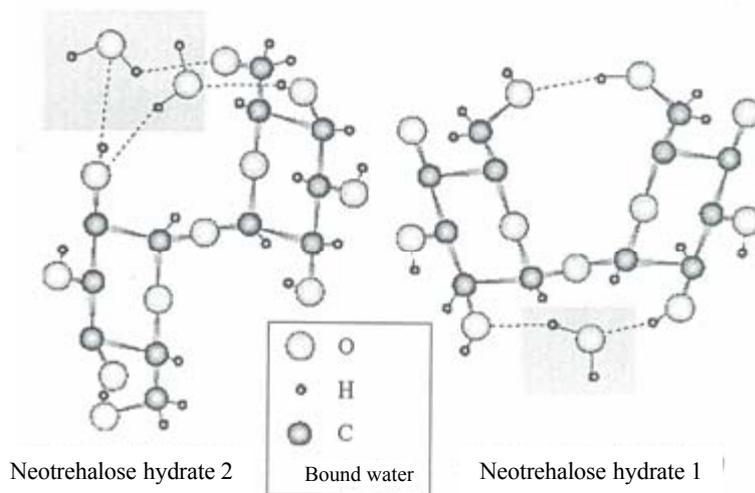


Fig. 1 Structures of trehalose dihydrate and neotrehalose hydrate

### Results and Discussions

Figure-2 shows IR spectra of trehalose dihydrate and neotrehalose hydrate at 68, 70, 72, 74 and 76°C in attribution to bound water. The peak of trehalose dihydrate in IR spectrum has shifted from 1680 cm<sup>-1</sup> to 1640 cm<sup>-1</sup> according to temperature rising, while the peak of neotrehalose has not. Though, maltose hydrate was measured by the same method, the result was the same as neotrehalose. Since it is said that the peak of ice is at 1703 cm<sup>-1</sup> and that of water, at 1640 cm<sup>-1</sup> generally, it can be considered that bound water of trehalose dihydrate was transformed the structure from ice-like to water-like in the range of 68–76°C. The melting point of trehalose is 96°C and trehalose dihydrate does not have intramolecular hydrogen bond unlike other disaccharides (Figure 1). Therefore, the structure transformation of bound water in the range of 68-76°C can be assumed as intramolecular structure is transformed while maintaining the crystal structure to some extent, and accordingly bound water is transformed from ice-like to water-like. Moreover it is estimated that the binding site of bound water was transformed very close to the structure of water along with the structure transformation. As a result, it is indicated that the binding site of this bound water may protect biological molecules as water mimic. It is considered that FTIR with its high sensitivity against water is a useful tool for the study in the structure of water of extremely small amount.

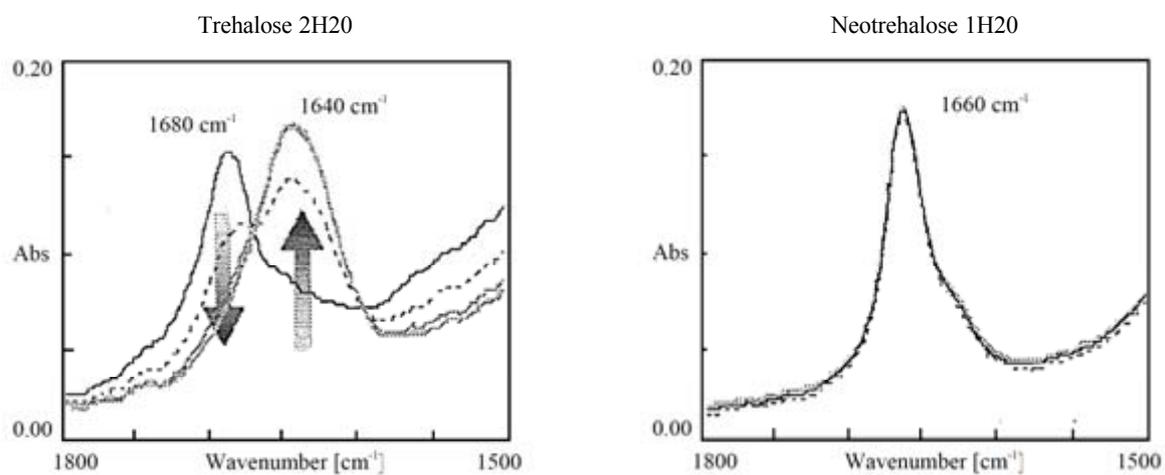


Fig. 2 IR spectra of trehalose dihydrate and neotrehalose hydrate in each temperature

#### Reference

K. Akao, Y. Okubo, T. Ikeda, Y. Inoue and M. Sakurai, *Chem. Letter*, 759 (1998)