Single Crystal X-ray Structural Analysis and Electronic Theory of Sugar Molecules

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Abstract. Rare sugar is a monosaccharide that there are trace amounts in nature, natural sugars present in large amounts in nature world is referred to as a naturally monosaccharide. Recently, the isomerase called D-tagatose-3-epimerase (DTE) was discovered, and it has a possibility to synthesize quantity of the rare sugar from a natural sugar through the enzymatic reaction. And all of hexoses can be synthesized by four kinds of enzymatic reactions (oxidoreductase, aldose isomerase, aldose reductase and DTE reactions). In this study, single-crystal X-ray structure analysis of a sugar alcohol and its derivative have been performed in order to determine the absolute coordinates of these sugar molecules. We have also investigated the electronic state calculation by means of DV-Xα method using the obtained data of the absolute coordinates. We also consider that mechanism of the enzymatic reaction and intermolecular energy properly such as a hydrogen bond in order to synthesize the supramolecular rare sugars (SRSs) which can be controlled structures and the hydrogen bonds.

1. Introduction.

1.1. Rare sugars and supramolecular rare sugars (SRSs).

Sugar is a kind of carbohydrate and an indispensable substance for most living organisms living on the earth. Sugars are broadly classified into monosaccharides and polysaccharides. A monosaccharide is a sugar that cannot be further hydrolyzed, and a polysaccharide refers to a monosaccharide linked by a glycosidic bond. Among the polysaccharides, two monosaccharides with glycosidic bonds are called disaccharides, three monosaccharides with glycosidic bonds are called trisaccharides, and ten to twenty monosaccharides with glycosidic bonds are called oligosaccharides.

Monosaccharides are finely classified according to their carbon number and structure, have aldehyde groups or ketone groups. As shown in Fig. 1, aldoses are those with an aldehyde group attached to the end of a monosaccharide chain, and ketoses are those where the second carbon is a ketone group. Further, a product obtained by reducing these aldehyde groups and ketone groups is called a sugar alcohol. A typical example of aldose is D-Glucose, a typical example of ketose is D-Fructose, and a typical example of sugar alcohol is D-Sorbitol. Although rare sugars are present in low amounts, there are more than 50 types of derivatives. Since rare sugars are rare and very expensive, they have not been studied much, and many of their properties such as structure, optical activity, and
physiological activity remain unknown. However, in recent years, D-fructose, a natural monosaccharide, was discovered by the discovery of an enzyme that catalyzes the reversible epithelization of the third carbon of ketose, called D-tagatose-3-epimerase (DTE), by Prof. Izumori Kagawa University, that can be converted to D-psicose, a rare sugar. Furthermore, the discovery of the DTE devised Izumoring, which systematizes enzyme reactions of various monosaccharides in a ring shape, enabling the strategic synthesis of the monosaccharides. Therefore, the main focus of this study is to elucidate the physical properties of the rare sugars located on the map of the Izumoring strategy.

![Figure 1](image1.png)

**Figure. 1** Synthetic strategy of hexose by enzymatic reaction (Izumoring).

A supramolecule is a molecule in which multiple molecules are assembled in an orderly manner by bonds other than covalent bonds, that is, hydrogen bonds or hydrophobic interactions, electrostatic interactions (Coulomb interaction), van der Waals interaction, etc. The concept of the supramolecules was proposed by Jean-Marie Lehn et al. Initially, compounds that encapsulate molecules and ions through intermolecular interactions, such as crown ethers and cyclodextrins, were particularly studied. Supramolecular rare sugar is an ordered arrangement of two or more different rare sugar molecules in a single unit cell, named by Professor Izumori.

In other words, supramolecular rare sugars are different from polysaccharides in which monosaccharides are glycoside-bonded molecules in which different molecules. They are regularly arranged with a relatively weak force due to hydrogen bonding. For this reason, it is easier to control the binding than polysaccharides, and it is considered that engineering applications such as an optical devices are easier.

**Single crystal X-ray structural analysis of rare sugars.**

In this study, crystal structure analysis is performed using an X-ray diffractometer. In 1895, X-rays were discovered by German physicist Wilhelm Conrad Röntgen, in 1912 Max Theodor Felix von Laue discovered the X-ray diffraction phenomenon. The discovery of Bragg’s law by William Henry Bragg and William Lawrence Bragg established the theory of the structural analysis using X-ray diffraction. In 1916, Peter Joseph William Debye and Paul Scherrer developed the Debye-Scherrer method, which can perform structural analysis from powder crystals, and structural analysis using X-ray diffraction became active. X-rays have very strong energy and are capable of passing through materials, so they are also being used in the field of nondestructive inspection. From the 20th century, it began to be used in fields such as structural biology.

Many analytical instruments that use X-ray diffraction phenomena use Bragg’s conditions. Many substances are made of extremely small crystals, and atoms and ions are regularly arranged inside the crystal to form a spatial lattice (unit cell). The spatial lattice has atoms arranged in parallel to each other, and when an X-ray with a wavelength \(\lambda\) is incident on the lattice plane with a surface interval \(d\) at an incident angle \(\theta\),

\[2dsin\theta = n\lambda \quad (n = 1,2,3\cdots).\]

The following relational expression holds. This is called Bragg’s condition. When this conditional expression holds, X-rays interfere with each other and strengthen each other.
2. The purpose of this study.
Since research on rare sugars has recently started full-scale research, many physical properties such as the structure and properties of rare sugars are unknown. The purpose of this study is to find applicability of rare sugars in the field of materials. In order to achieve this goal, it is necessary to elucidate the chemical and physical properties of rare sugars in detail. Moreover, intermolecular interactions play an important role in designing organic materials, and the molecular structure often affects this interaction. Therefore, the molecular structure of rare sugars and the high-precision absolute coordinates of each atom are indispensable for future research on rare sugars. The purpose of this study is to investigate the mechanism of the hydrogen bonding network and optical activity of sugars by analyzing the crystal structure and electronic state of rare sugars, and to synthesis the supramolecular rare sugars.

3. Experimental Method.
In this study, a single crystal obtained by precipitating a 15 % concentration of Galactitol solution (mixed solution of 1.5 g of Galactitol and 10 mL of water) at 60 °C for 24 hours, and an Allitol solution of 35 % concentration (Allitol 1.05 g). A single crystal obtained by precipitating a mixed solution of 3.0 mL of water at 30 °C for 48 hours is subjected to crystal diffraction using an X-ray structural analyzer R-AXIS RAPID II (Rigaku Corporation). The diffraction data was subjected to structural analysis using structural analysis software (Crystal Structure). Furthermore, using the obtained crystal structural data, BOP (Bond Overlap Population), HOMO-LUMO gap, total energies of Galactitol and Allitol were calculated by using DV-Xα method. Besides we calculated four sugars (D-Fructose, D-Psicose, D-Galactose and D-Glucose) by using the obtained crystal structure data and derive these data to consider four kinds of enzymatic reactions which synthesize all of hexoses (oxidoreductase, aldose isomerase, aldose reductase and DTE reactions).

4. Results and Discussion.
4.1. Single crystal X-ray structural analysis of Galactitol and Allitol.
First, we have obtained more precise single crystal structure of Galactitol and Allitol. These polyols have no distinction between D and L forms and do not have an optical isomers. Since these molecules are located at the conversion points of D-form and L-form in the Izumoring strategy, they can be said to be important molecules among Izumoring molecules.

Figure. 2 Crystal structure of Galactitol.

Figure. 3 Crystal structure of Allitol.
Table 1 Cell parameters for Galactitol and Allitol.

|                  | Galactitol | Allitol |
|------------------|------------|---------|
| Formula          | C₈H₁₂O₆    | C₆H₁₄O₆ |
| Crystal System   | monoclinic | monoclinic |
| Space Group      | P₂₁/c      | P₂₁     |
| Cell Length [Å]  | a 8.4051(6) | 4.6873(4) |
|                  | b 11.4534(8) | 13.3361(10) |
|                  | c 9.0027(6) | 6.5887(5) |
| Cell Angles [°]  | α 90       | 90      |
|                  | β 112.995(8) | 100.027(7) |
|                  | γ 90       | 90      |
| Cell Volume [Å³] | 797.892   | 405.56(5) |
| Z                | 4          | 2       |
| R-Factor [%]     | 4.01       | 7.03    |

Figs. 2 and 3 show the crystal structures of Galactitol and Allitol obtained by the crystal structural analysis, and Table 1 shows their cell parameters. In Figs. 2 and 3, the red, green, and blue lines indicate the a, b, and c-axis of the unit cell, respectively, and each molecule is displayed in the Ball and Stick format. One molecule of Galactitol was confirmed to be present in the unit cell by four molecules by the symmetry operation of the space group P₂₁/c. In Figs. 2 and 3, the twice (2₁ symmetric) helical axis extends in the direction parallel to the b-axis, and the Galactitol molecule has a projection plane perpendicular to the double helical axis, the ab-axis plane (c-axis direction) is exist parallel to the axial direction. The R values obtained from this analysis result, which are the standard for the accuracy of the structural analysis results, were 4.01 % and 7.03 %, and high-precision absolute coordinates were obtained.

4.2. Electronic state calculation of main hexoses.

Second, we calculated two polyols (Galactitol and Allitol), two ketoses (D-Fructose and D-Psicose) and two aldoses (D-Galactose and D-Glucose) by using the obtained crystal structure data (Table 2). Comparing Galactitol and Allitol, the BOP around C₂ is generally larger for Galactitol, and even when comparing the symmetry and density of the space group, Galactitol is higher, and the crystallinity of the whole molecule is better than Allitol.

Comparing D-Glucose and D-Fructose, HOMO-LUMO gap is larger for D-Fructose (9.600 eV) and the BOP around C₂ is roughly larger for D-Fructose. In nature, D-Glucose is predominantly present in natural monosaccharides. It is shown that the lower the value of the parameter (BOP and HOMO-LUMO gap) related to intramolecular stability, the more it can exist in nature. This is thought to be the fact that the higher the intramolecular stability, the lower the stability of the crystal, leading to a decrease in the abundance in nature.
Table 2. Structure data, BOP, HOMO-LUMO gap and Total Energy of main hexoses (Galactitol, Allitol, D-Fructose, D-Psicose, D-Galactose and D-Glucose).

|          | Space Group | Density (g/cm³) | BOP C2-H | C2-C1 | C2-OH | C2-C3 | HOMO-LUMO (eV) | Total Energy (Hr) | Cavity (%) |
|----------|-------------|----------------|----------|-------|-------|-------|---------------|------------------|------------|
| Galactitol | P2₁/c       | 1.516          | 0.8121   | 0.7629| 0.5728| 0.7583| 8.4399        | -680.94         | 11.1       |
| Allitol   | P2₁         | 1.472          | 0.8080   | 0.7392| 0.5849| 0.7392| 9.4880        | -680.72         | 10.8       |
| D-Fructose| P2₁,2,2₁    | 1.602          | 0.7945   | 0.7702| 0.5812| 0.7531| 9.600         | -679.49         | 10.8       |
| D-Psicose | P2₁,2,2₁    | 1.606          | 0.8166   | 0.7459| 0.5853| 0.7587| 9.447         | -679.41         | 11.8       |
| D-Galactose| P2₁,2,2₁   | 1.580          | 0.8228   | 0.7404| 0.5821| 0.7476| 9.405        | -679.69         | 12.3       |
| D-Glucose | P2₁,2,2₁    | 1.575          | 0.7801   | 0.7316| 0.5045| 0.7536| 8.864         | -679.40         | 11.8       |

5. Conclusion
In the crystal structure analysis of Galactitol and Allitol, the R value was improved by succeeding in identifying the position of the hydrogen atom considering the direction of intermolecular hydrogen bonding, and the crystal structure that exists more stable than the crystal structure on the CSD Succeeded in finding out. In addition, more accurate electronic state calculations related to intermolecular interactions are possible. Galactitol and Allitol are the central molecules in Izumoring, and they have the common feature of sugar alcohols located at the conversion points of D-forms and L-forms. In addition, parameters related to the stability of molecules and crystals could be obtained by calculating the electronic structure using precise crystal structure data. The data obtained by these electronic state calculations will be the most useful means for elucidating the principles, phenomena and principles in nature.

References
[1] F. Longchambon, H. Gillier-Pandraud, R. Wiest, B. Rees, A. Mitschler, R. Feld, M. Lehmann, P. Becker, Acta Crystallogr. Sect. B: Struct. Sci., 41, 47, (1985).
[2] K. Fukada, T. Ishii, K. Tanaka, M. Yamaji, Y. Yamaoka, K. Kobashi, K. Izumori, Bull. Chem. Soc. Jpn., 83, 1193, (2010).