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Complexation of Cycloinulohexaose with Some Metal Ions

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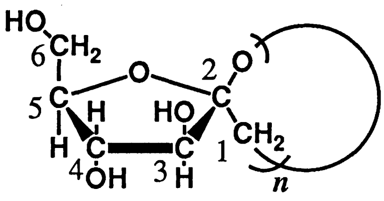
Cycloinulohexaose(CF6), which is a cyclohexaose composed of six(2⟶1) linked β-D-fructofuranosyl units, has a 18-crown-6 skeleton in its center. Selective complexation of CF6 with some metal ions have been observed in D_2O by NMR spectroscopy. The variation of ^1H NMR chemical shifts indicates that the ether oxygen atoms of CF6 should act as electron donors, which interact electrostatically with the metal ions.

Cyclofructans(CFs,**1**), which are produced from inulin by using the extracellular cycloinulo-oligosaccharide fructanotransferase from Bacillus circulans MCI-2254,$^{1)}$ are cyclo-oligosaccharides composed of at least six (2⟶1) linked β-D-fructofuranosyl units. In CFs, (2⟶1) glycosidic linkages constitute a "crown ether-like" skeleton, i.e. $\overbrace{(\text{C-C-O})}_n$. CFs are distinct from typical synthetic crown ethers in their three characteristics: (1) the ether ring of CFs is surrounded by hydrophilic groups of fructofranose residues. (2) The internal motion of the ring should be restricted because of the large substituents. (3) Conformational arrangement of the ring in the crystalline state is different from typical synthetic crown ethers. The



1 (n=6,7,8)

ether ring of CF6(cycloinulohexaose) has a gtgtgt conformation around the $-C-CH_2-$ bonds in the uncomplexed crystalline state,[2)] whereas the arrangement is $\mathrm{gt\bar{g}\bar{g}tg}$ for the uncomplexed state of a simple crown ether with the same ring size, i.e., 18-crown-6[3)] and $\mathrm{g\bar{g}g\bar{g}g\bar{g}}$ for its typical complexes with metal ions such as K^+ [4)] and Cs^+.[5)] Thus, CFs should have potential for forming new type complexes with metal cations. In this letter, we report the first observation of selective complexation of CF6 with some metal ions in D_2O by NMR spectroscopy.[6)]

Figure 1 shows 270 MHz ^1H NMR spectra of free and complexed CF6 in D_2O.[7)] For free CF6, the peak assignments were performed by measurement of a ^1H-^1H COSY NMR spectrum. The assignments are shown in Fig. 1(a). For CF6 in the crystalline state, six furanose residues are alternately above and below the mean plane of the ether ring.[2)] Two kinds of nonequivalent furanose residues exist in this state. The NMR spectrum of CF6 in solution, however, shows the existence of only one kind of residues. Thus, the furanose residues should be perturbed up and down toward the plane of the ether ring in the solution. This perturbation is faster than the chemical shift difference of the two nonequivalent residues.

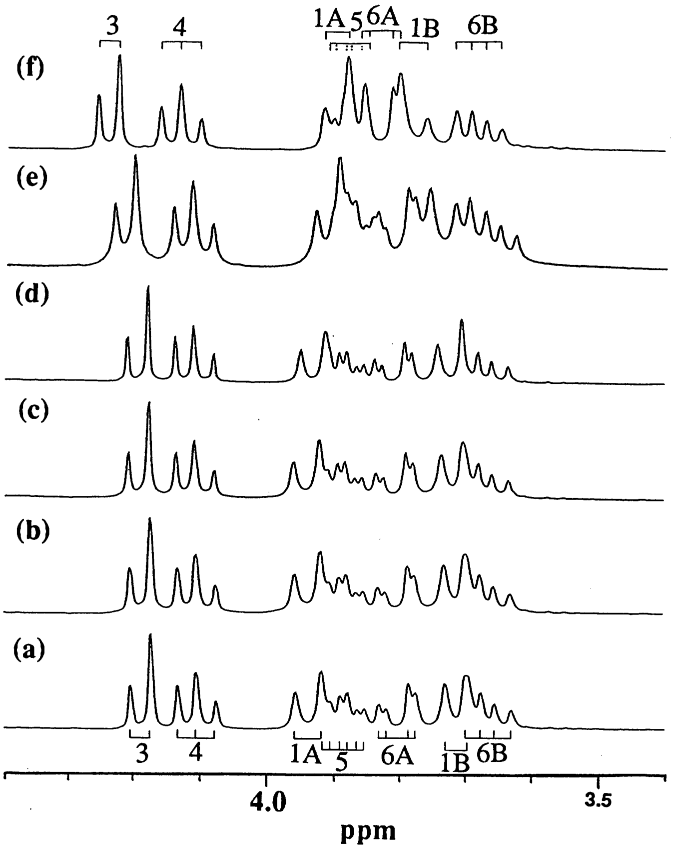


Fig.1. ^1H NMR spectra of uncomplexed and complexed CF6 in D_2O: (a) CF6; (b) CF6 + $CaCl_2$; (c) CF6 + NaCl; (d) CF6 + KCl; (e) CF6 + $PbCl_2$; (f) CF6 + $BaCl_2$. The concentrations of CF6 and metal ions were 13 and 30 mM, respectively.

When metal ions were added

to CF6 solutions, ^1H NMR spectra were varied from that of the free state (Fig. 1(b)-(f)). The effects were significant for Ba^{2+} and Pb^{2+}. The small effects were also observed for K^+. No shifts were observed for Na^+ and Ca^{2+}. Since all of these solutions contain Cl^- anion with concentration of 30 or 60 mM, these shifts did not result from ther influence of Cl^-. Under these experimental conditions, complexations of CF6 with Ba^{2+}, Pb^{2+}, and K^+ were clearly indicated.

In the spectra of the three complexes, the H3, H4, and H1B resonances shift downfield considerably. Only the H1A resonance shifts upfield. The shift for H5 and H6 scarcely change. This observation indicates that the metal ions should be positioned near the ether ring of CF6. The ether oxygen atoms of CF6, as well as those of 18-crown-6, should act as electron donors, which interact with the metal ions electrostatically. In this experimmental conditions, the resonances from hydroxyl groups cannot observed. Thus, it is not clear whether the hydroxyl oxygen atoms act as electron donors or not. This is the first observation that the macrocyclic oligosaccharide forms crown ether-like complexes with some metal ions.

The changes in the chemical shifts of CF6 were measured as a function of increasing metal ion/CF6 ratios. The dissociation constant K_d ($=[CF6]\cdot[Metal^+]/[CF6\cdot Metal])^+$ of CF6 with Ba^{2+}, Pb^{2+}, and K^+ were determined by Benesi-Hildebrand treatment[8)] using a simple equilibrium model of $CF6 + Metal^+ \longrightarrow CF6\cdot Metal^+$. The CF6-$Ba^{2+}$, CF6-$Pb^{2+}$, and CF6-$K^+$ complexes have K_d's of $4.6*10^{-2}$, $5.6*10^{-2}$, and $4.1*10^{-1}$M, respectively. The complexation constant of 18-crown-6 is dependent on the ion radius and is large for a metal ion having a radius compatible with the size of the 18-crown-6 cavity. The K_d's between 18-crown-6 and metal ions are reported to be in order of $Pb^{2+} < Ba^{2+} << K^+ << Na^+ < Ca^{2+}$ in H_2O.[9)] The same order of the K_d was suggested for CF6. The ion selectivity should also depend on the same factors as typical synthetic crown ethers.

Further NMR study is now in progress for determination of the structures of the complexes and the interaction of other CFs and modified CFs with metal ions.

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6) CF6 was courteously supplied from Mitsubishi Kasei Corporation.
7) ^{1}H NMR spectra (270 MHz) were recorded on a JEOL GSX-270 spectrometer at 35 °C in D_2O. The concentration of CF6 and metal ions were 13 and 30 mM, respectively.
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