

ARTICLES

Calculations of Molecular Surface Area Changes with Docking of Host and Guest and Applications to Cyclodextrin Inclusion

Seiji Ishikawa, Sakae Hada, Saburo Neya, and Noriaki Funasaki*

Kyoto Pharmaceutical University, Misasagi, Yamashina-ku, Kyoto 607-8414, Japan

Received: July 29, 1998; In Final Form: November 4, 1998

A method for calculations of molecular surface area changes with the docking of host and guest is developed and applied to the estimation of the structures and binding constants of cyclodextrin inclusion systems. Each molecule of the host and guest is regarded to consist of hydrophilic and hydrophobic groups. The change ΔS in water-accessible surface area with the docking of these host and guest molecules is divided into four terms: $\Delta S_{oo}(HG)$, $\Delta S_{ow}(HG)$, $\Delta S_{wo}(HG)$, and $\Delta S_{ww}(HG)$. For instance, $\Delta S_{oo}(HG)$ stands for the change in host hydrophobic surface area by overlapping with guest hydrophobic surface area. When a guest molecule is moved along the symmetry axis of cyclodextrin, the structure of the complex having the maximum $\Delta S_{oo}(HG)$ value is close to its crystal structure. Thus, we can estimate the "solution" structure of the complex from the maximum $\Delta S_{oo}(HG)$ value. Using this method, we predict the solution structures of six cyclodextrin inclusion systems. Furthermore, we find that the logarithm of the 1:1 binding constant is linear with the maximum $\Delta S_{oo}(HG)$ value for 11 systems including α -, β -, and γ -cyclodextrins and aliphatic and aromatic guest molecules. The present results would be applied to other cyclodextrin inclusion systems and protein–ligand systems.

Introduction

Cyclodextrins (CyDs) have homogeneous toroidal structures of different cavity sizes. Three of the most characterized CyDs are α -, β -, and γ -CyDs, which contain six, seven, and eight glucose units, respectively. The toroidal structure has a hydrophilic surface resulting from the 2-, 3-, and 6-position hydroxyls, making CyD water-soluble. Its cavity is composed of the glucoside oxygens and methylene hydrogens, giving it a hydrophobic character. As a consequence, CyDs can include other hydrophobic molecules of appropriate dimensions inside their cavity. To a first approximation, the magnitude of binding constants correlates with the fit of the guest in the CyD cavity. Therefore, CyDs can give beneficial modifications of guest molecules not otherwise achievable: solubility enhancement, stabilization of labile guests, control of volatility and sublimation, and physical isolation of incompatible compounds. Because they are practically nontoxic, they are added into pharmaceuticals and foods.^{1,2}

In some reviews and books, the data on the crystal^{3,4} and solution structures of CyD complexes and the binding constants are summarized and several driving forces of CyD complexation are suggested.^{1–5} These forces include CyD ring strain and van der Waals forces, hydrophobic interactions, and hydrogen bonds between CyD and guest. Such driving forces of complexation, despite the many papers dedicated to this problem, have not yet been understood fully.^{1–5} Molecular-mechanical and molecular dynamic calculations have been applied to estimate the structures of complexes.⁶

The included molecules are normally oriented in the host in such a position as to achieve the maximum contact between the hydrophobic part of the guest and the hydrophobic CyD cavity. The hydrophilic part of the guest molecule remains, as far as possible, at the outer face of the complex. This ensures maximum contact with both the solvent and the hydroxyl groups of the host.² For this approach, we must calculate molecular surface areas of contact between host and guest. To our knowledge, there is no method for calculations of such overlapped surface areas. Connors investigated the correlation between the changes in molecular surface area and the binding constants for complexations of many guests with α -CyD.^{5,7} Using water-accessible surface area data of complexes of β -CyD with methyl-substituted phenols, Mayer et al. theoretically analyzed their three-dimensional structures.⁸

Although the physical picture of hydrophobic interactions is still unclear, the magnitude of hydrophobicity or hydration free energy of a solute is empirically linear with its water-accessible surface area.^{9–12} This relationship is widely used to analyze aqueous solubility,^{9,13} water/oil partition coefficients,^{13–15} critical micelle concentrations,^{16–18} capacity factors in reversed phase HPLC,^{19,20} and biological activity²¹ of small molecules as well as the unfolding^{11,22} and binding^{23–26} of proteins.

Analytical^{9,13} and numerical¹⁸ methods for calculating molecular surface areas are used in the literature.²³ In this work we have extended our numerical method¹⁸ for calculating hidden molecular surface areas of complexes and have applied this method to predict stable structures of CyD complexes and their binding constants. The present method should be applicable to other host–guest systems.²⁷

* Fax: +81-75-595-4762. E-mail: funasaki@mb.kyoto-phu.ac.jp.

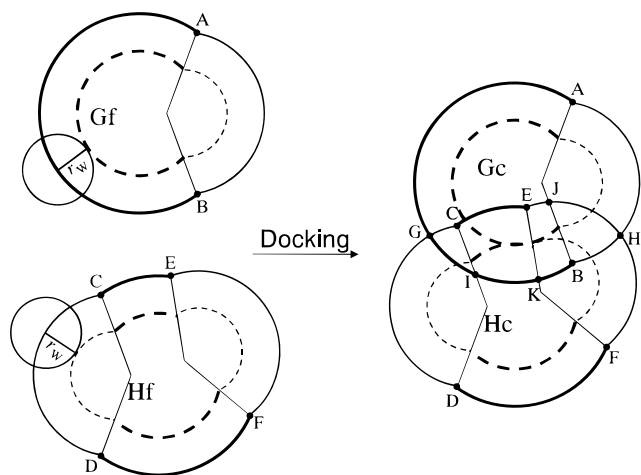


Figure 1. Definitions of hydrophilic and hydrophobic surface areas for free guest (Gf), free host (Hf), and complex (HG). The hydrophilic surface is shown in thin lines and the hydrophobic surface in thick lines. The dashed lines denote the van der Waals surface, and the solid lines depict the water-accessible surface. For the complex, each arc constitutes part of the following area: arc AG, $So(Gc)$; arc AH, $Sw(Gc)$; arcs BK and GI, $\Delta S_{ow}(Gc)$; arc IK, $\Delta S_{oo}(Gc)$; arc BH, $\Delta S_{ww}(Gc)$; arc DF, $So(Hc)$; arcs DG and FH, $Sw(Hc)$; arc CE, $\Delta S_{oo}(Hc)$; arcs CG and EJ, $\Delta S_{wo}(Hc)$; arc HJ, $\Delta S_{ww}(Hc)$.

Theory

Definitions of Changes in Molecular Surface Area with Docking. Water-accessible molecular surface area S was defined as the area of surface traced out by the center of a water molecule rolling over the van der Waals surface of the solute molecule (Figure 1).⁹ The solute molecule consists of hydrophilic atoms and hydrophobic (oleophilic) atoms. Thus, the molecular area S consists of hydrophilic area Sw and hydrophobic area So . This definition is applicable to the equimolar complex (HG) of host (H) and guest (G). The surface area of the complex consists of hydrophilic and hydrophobic parts, viz., $S(HG) = Sw(HG) + So(HG)$. This area is also split into contributions of the host and guest, viz., $S(HG) = S(Hc) + S(Gc)$. The hydrophobic part of $So(HG)$ is equal to $So(Hc) + So(Gc)$. The definitions of other surface areas are summarized in the Nomenclature.

Next, let us consider the change in surface area with the docking of host and guest under the assumption that their molecular conformations do not change (Figure 1). Then the decrease in S with the docking is written as

$$\Delta S(HG) = S(Hf) + S(Gf) - S(HG) \quad (1)$$

This decrease consists of the contributions of the host and guest, viz., $\Delta S(Hc) + \Delta S(Gc)$. The decrease in hydrophobic area of host with the docking can be written as

$$\Delta So(Hc) = So(Hf) - So(Hc) \quad (2)$$

Similarly, we can define $\Delta So(Gc)$, $\Delta Sw(Hc)$, and $\Delta Sw(Gc)$.

The decreased hydrophobic area of host, $\Delta So(Hc)$, consists of $\Delta S_{oo}(Hc)$ and $\Delta S_{ow}(Hc)$: The former area denotes part of the hydrophobic host surface in complex in contact with hydrophobic atoms of the guest and the latter stands for that in contact with hydrophilic atoms of guest. Furthermore, we can also define $\Delta S_{oo}(Gc)$, $\Delta S_{ow}(Gc)$, $\Delta S_{wo}(Gc)$, $\Delta S_{ww}(Gc)$, $\Delta S_{wo}(Hc)$, and $\Delta S_{ww}(Hc)$ for the host and guest in complex (see Nomenclature). These will be termed hidden water-accessible surface areas. When host and guest form a complex,

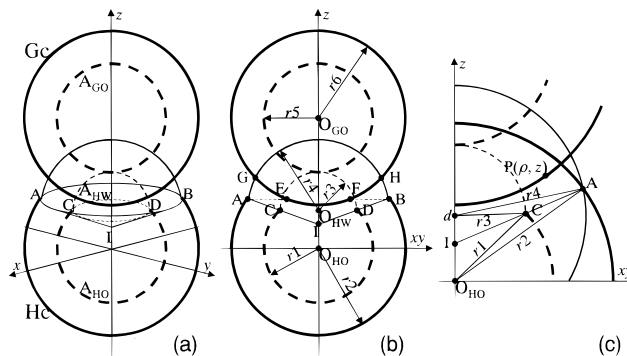


Figure 2. Boundary between different water-accessible surfaces of the host molecule composed of two hydrophilic and hydrophobic atoms, where its hydrophilic atom is in the van der Waals contact with the hydrophobic atom of the guest. (a) Three-dimensional view of the complex molecule composed of three atoms whose centers (O_{HO} , O_{HW} , and O_G) are located on the z axis. (b) Cross-section of Figure 2a by a plane passing the z axis. (c) Partial expansion of the overlapping region in Figure 2b. Here d = distance between the atomic centers of the host molecule, r_1 = van der Waals radius of the hydrophobic atom of the host, $r_2 = r_1 + r_w$, r_3 = van der Waals radius of the hydrophilic atom, and $r_4 = r_3 + r_w$. The Cartesian coordinate centered at the hydrophobic atom of the host is based on the intramolecular coordinate for the host molecule. In Figure 2b points E and F are the intersections of line AB and the water-accessible surface of the guest molecule. In Figure 2c point I is defined as the intersection of line AC and the z axis, and point P can move on the water-accessible surface of the guest molecule.

these molecules can approach their van der Waals surfaces beyond their water accessible surfaces.

In Figure 1, the thick solid line displays the hydrophobic water-accessible surface and the thin dashed line denotes the hydrophilic van der Waals surface. The guest molecule is composed of a hydrophilic atom and a hydrophobic atom, and the host consists of two hydrophilic atoms and a hydrophobic atom. For example, the thick arc AB of free guest (Gf) constitutes part of $So(Gf)$. When the guest forms the complex with the host, arc AB(Gf) becomes arcs AG(Gc), GI(Gc), IK(Gc), and KB(Gc). The thick arc AG(Gc) is part of $So(Gc)$, because it does not overlap with the host. Because arc IK(Gc) is contained in the hydrophobic volume of the host, it becomes part of $\Delta S_{oo}(Gc)$. Arcs GI(Gc) and KB(Gc) are parts of $\Delta S_{ow}(Gc)$. The thin arc EF(Hf) becomes arcs EJ(Hc), JH(Hc), and HF(Hc), when the host complexes with the guest. The first arc is part of $\Delta S_{wo}(Hc)$, the second is part of $\Delta S_{ww}(Hc)$, and the last is part of $Sw(Hc)$.

Boundary between Hydrophilic and Hydrophobic Surfaces in Complex. To calculate all of the hidden surface areas defined above for the complex, we must decide which part of each atom of the guest molecule is in contact with hydrophilic or hydrophobic atoms of the host molecule and vice versa.

To illustrate this problem, we assume a special complex molecule composed of three atoms (Figure 2), where the atoms are all located on the z axis and the hydrophilic atom (named A_{HW}) of host is in van der Waals contact with the hydrophobic atom (named A_{GO}) of guest. The centers of A_{HW} and A_{GO} are denoted by O_{HW} and O_{GO} , respectively. The water-accessible surfaces of these atoms overlap when

$$\text{distance } O_{GO}O_{HW} < r_4 + r_6 \quad (3)$$

The water-accessible surfaces of A_{GO} and A_{HO} (centered at O_{HO}) overlap, when

$$\text{distance } O_{GO}O_{HO} < r_2 + r_6 \quad (4)$$

The position vectors, $\mathbf{O}_{\text{HO}}[x_{\text{HO}}, y_{\text{HO}}, z_{\text{HO}}]$, $\mathbf{O}_{\text{HW}}[x_{\text{HW}}, y_{\text{HW}}, z_{\text{HW}}]$, and $\mathbf{O}_{\text{GO}}[x_{\text{GO}}, y_{\text{GO}}, z_{\text{GO}}]$, represent the centers of A_{HO} , A_{HW} , and A_{GO} , respectively. Then the z axis is written as $[\mathbf{O}_{\text{HO}} + t(\mathbf{O}_{\text{HW}} - \mathbf{O}_{\text{HO}})]$, where t stands for a variable parameter. The interatomic distance, d , between A_{HW} and A_{HO} must satisfy the following inequalities:

$$|r_1 - r_3| < d < r_1 + r_3 \quad (5)$$

The hydrophobic water-accessible surface area, $So(\text{Gf})$, of the guest molecule is composed of $So(\text{Gc})$, $\Delta\text{Soo}(\text{Gc})$, and $\Delta\text{Sow}(\text{Gc})$ by docking with the host molecule. The $So(\text{Gc})$ value is easily calculable in a similar manner as was made for a free molecule.¹⁸ To separate the rest of $[So(\text{Gf}) - So(\text{Gc})]$ into $\Delta\text{Soo}(\text{Gc})$ and $\Delta\text{Sow}(\text{Gc})$, we must determine the boundary between these hidden areas.

We generate point P on the water-accessible surface of the guest molecule and examine how close this point is to the host molecule. In the world coordinate system, point P is represented by the following position vector:

$$\mathbf{P}_w[wx, wy, wz] = \mathbf{O}_{\text{GO}}[x_{\text{GO}}, y_{\text{GO}}, z_{\text{GO}}] + \Phi[r_6, \theta, \phi] \quad (6)$$

Here Φ is a well-known operator of transformation from spherical polar coordinates to Cartesian coordinates.¹⁸ As Figure 2a shows, we can consider two cones for the host molecule. The first cone has bottom circle AB, which is the plane of contact between the water-accessible surfaces of A_{HO} and A_{HW} . Circle AB satisfies the following equations:

$$z_1 = \{(r_2^2 - r_4^2)/d + d\}/2 \quad (7)$$

$$x^2 + y^2 = r_2^2 - z_1^2 \quad (8)$$

The second cone has bottom circle CD, which is the plane of contact between the van der Waals surfaces of these atoms. This circle CD satisfies the following equations:

$$z_2 = \{(r_1^2 - r_3^2)/d + d\}/2 \quad (9)$$

$$x^2 + y^2 = r_1^2 - z_2^2 \quad (10)$$

These cones IAB and ICD have the common top shown by point I and are shown two-dimensionally as two isosceles triangles, ΔABI and ΔCDI , in Figure 2b.

Figure 2c shows a partial expansion of the overlapping region in the two-dimensional system. Here the ρ coordinates (a representative of the x and y coordinates) of points A and C can be written as

$$\rho_2 = (r_2^2 - z_2^2)^{1/2} \quad (11)$$

$$\rho_1 = (r_1^2 - z_1^2)^{1/2} \quad (12)$$

In this ρ - z coordinate system we define the following function:

$$f(\rho, z) = (\rho_1 z_2 - \rho_2 z_1) \{ (\rho - \rho_1)(z_2 - z_1) - (z - z_1)(\rho_2 - \rho_1) \} \quad (13)$$

Line AC satisfies the equality

$$f(\rho, z) = 0 \quad (14)$$

Point I is the z section of line AC. This line indicates the border between A_{HO} and A_{HW} . Namely, when point P is present below this line $[f(\rho, z) < 0]$, it falls within the water-accessible volume of A_{HO} . When this point lies above the line $[f(\rho, z) > 0]$, it falls

within the water-accessible volume of A_{HW} . In the present special case, because point P belongs to the upper region, arc GEFH is part of $\Delta\text{Sow}(\text{Gc})$ and $\Delta\text{Soo}(\text{Gc})$ is null.

We have just considered the boundary problem in the two-dimensional coordinate system (Figure 2c). Next, we must consider this problem in the three-dimensional coordinate system (Figure 2a). The boundary is represented by cone ABI. We can examine which atomic region of the host molecule contains with point P as follows. First, we transform the absolute three-dimensional coordinates of the point to the intramolecular coordinates defined for the host molecule. This intramolecular coordinate system, centered at \mathbf{O}_{HO} , is written in Figure 2a. The vector of point P in this three-dimensional system can be written as

$$\mathbf{P}_m[px, py, pz] = \mathbf{M}\mathbf{P}_w \quad (15)$$

where \mathbf{M} is the operator of transformation from world to intramolecular coordinates:

$$\mathbf{M} = \begin{pmatrix} y_m/\rho_m & -x_m/\rho_m & 0 \\ -x_m z_m/\rho_m d & -y_m z_m/\rho_m d & \rho_m/d \\ x_m/d & y_m/d & z_m/d \end{pmatrix} \times \begin{pmatrix} 1 - x_{\text{HO}}/wx & 0 & 0 \\ 0 & 1 - y_{\text{HO}}/wy & 0 \\ 0 & 0 & 1 - z_{\text{HO}}/wz \end{pmatrix} \quad (16)$$

Here

$$[x_m, y_m, z_m] = \mathbf{O}_{\text{HW}} - \mathbf{O}_{\text{HO}} \quad (17)$$

$$\rho_m = (x_m^2 + y_m^2)^{1/2} \quad (18)$$

We can therefore see the boundary symmetric with respect to the z axis as well as to the two atoms of the host molecule. Then we get the distance ρ between point P and the z axis to determine the two-dimensional intramolecular coordinates of point P:

$$\mathbf{P}(\rho, z) = \{[(px)^2 + (py)^2]^{1/2}, (pz)\} \quad (19)$$

After having made these symmetry operations, we can finally treat a three-dimensional problem as a two-dimensional one and need not take into consideration negative ρ values. The above definition of the intra-atomic boundary can be extended to more complicated complexes, although it is not easy to illustrate such cases two-dimensionally.

If we incorrectly employed line AEFB of Figure 2b as the boundary, arcs GE and FH and arc EF would constitute parts of $\Delta\text{Sow}(\text{Gc})$ and $\Delta\text{Soo}(\text{Gc})$, respectively. The $\Delta\text{Soo}(\text{Gc})$ value is not null. Thus, this is an inappropriate definition of the boundary.

Calculating Procedures. We have already reported a numerical method for calculations of water-accessible atomic surface areas.¹⁸ Our method is based on the counting of dots generated on the molecular surface. Each dot (point), e.g., point P in Figure 2c, is located at the center of a curved rectangle having a given area. Each atom is judged to be hydrophilic or hydrophobic, and its surface area is calculated. Summing up its area over all hydrophilic and hydrophobic atoms in the molecule, we can determine S_w and S_o for the molecule. This method is also applicable to the complex of host and guest without modifications. Figure 3a shows a flowchart for calculations of the exposed water-accessible surface areas of host and guest as well as their complex.

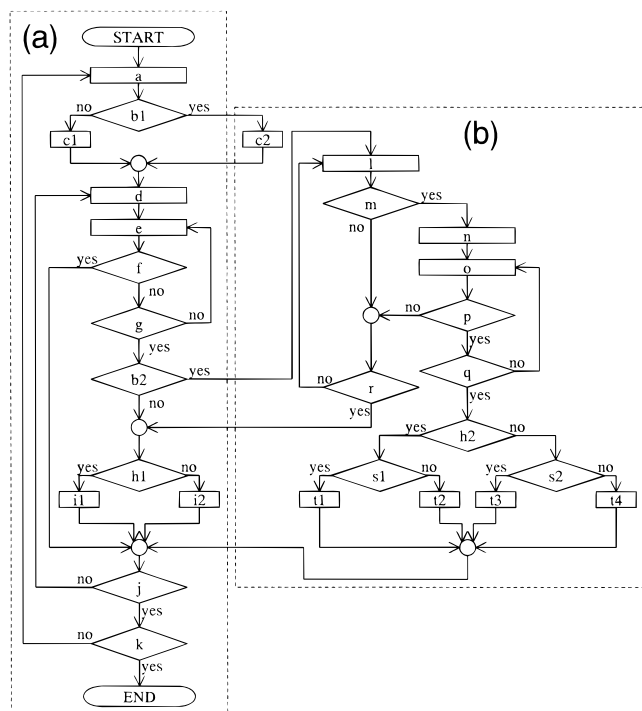


Figure 3. Flowchart for calculating molecular surface areas of a guest molecule in complex: (a) main chart and (b) subchart for hidden surface areas. a: Choice of atom A making up the host molecule. b1 and b2: Do you calculate the hidden surface area of the guest molecule? c1: List all atoms overlapping with atom A from host and guest molecules (List 1). c2: List all atoms overlapping with atom A from host (List 2) and guest (List 1) molecules. d: Production of the vector indicating the place of area element P on the sphere of atom A. e: Calculation of the distance D between the centers of P and atom L1, one of the atoms in List 1. f: $D < \text{radius of atom L1}$? g: Did you finish calculations over all overlapping atoms? h1 and h2: Is atom A hydrophilic? i1: Addition of area of P to $S_w(G)$. i2: Addition of area of P to $S_o(G)$. j: Did you finish all area elements of atom A? k: Did you finish steps a to j for all atoms constituting the guest molecule? l: Choice of atom B from List 2. m: Distance between centers of P and atom B $<$ the radius of atom B? n: Choice of all atoms overlapping with atom B from List 2 and addition of these atoms into List 3. o: Determination of the boundary between atom B and one of the atoms in List 3. p: Is area element P inside atom B? q: Did you finish steps o and p over all atoms in List 3? r: Did you finish steps m to p over all atoms in List 2? s1 and s2: Is atom B hydrophilic? t1: Summation of the area element to $\Delta S_{ww}(Gc)$. t2: Summation of the area element to $\Delta S_{wo}(Gc)$. t3: Summation of the area element to $\Delta S_{ow}(Gc)$. t4: Summation of the area element to $\Delta S_{oo}(Gc)$.

In this work we have extended this method to estimate water-accessible surface areas hidden by the contact of host and guest. Figure 3b shows a flowchart for calculating the hidden surface areas of their complex. Following this flowchart, we will demonstrate how to calculate the exposed and hidden water-accessible areas of the complex shown in Figure 2. We choose one (A_{GO}) of the atoms constituting the host molecule (step a) and investigate whether the water-accessible surface of this atom intersects the water-accessible surfaces of the other atoms of host and guest ("yes" at step b1). Because the guest is a monoatomic molecule, there is no guest atom overlapping with the water-accessible surface of A_{GO} (no guest atom in List 1). However, the water-accessible surface of A_{GO} intersects the water-accessible surfaces of both A_{HO} and A_{HW} , because eqs 3 and 4 are satisfied. These atoms HO and HW are included in List 2 (step c2). We generate point P on the water-accessible surface of A_{GO} at step d. Because there is no atom in List 1, we can skip steps e–g.

Next, we check the overlapping of point P with the water-accessible surfaces of the host atoms in List 2 ("yes" at step b2). At step l, A_{HO} is selected from these atoms. The point P lies in the volume of A_{HO} ("yes" at step m, viz., distance $PO_{HO} < r_2$). We must examine whether this point is contained in the water-accessible volume of A_{HO} . At step n we choose A_{HW} . Because this atom overlaps with the water-accessible sphere of A_{HO} , it is included in List 3. At step o, we determine the boundary between A_{HO} and A_{HW} using eqs 7–13. Because point P is not inside A_{HO} [$f(\rho, z) > 0$] at step p, this point is not hidden by the water-accessible surface of A_{HO} . Because this case is "no" at both steps p and r, we return to step l to investigate the overlapping of point P with the water-accessible volume of A_{HW} for the other atoms in List 2. Through steps m–p, the same procedures with A_{HO} are repeated for A_{HW} . Because this case is "yes" at steps p and q, point P is hidden by the water-accessible volume of A_{HW} . This area element, centered at point P, is summed up into $\Delta S_{ow}(Gc)$ at step t3, because A_{GO} is a hydrophobic atom ("no" at step h2) and A_{HW} is a hydrophilic atom ("yes" at step s2). In the case where point P is outside both the water-accessible volumes of A_{HO} and A_{HW} ("no" at step m and "yes" at step r, viz., distance $PO_{HO} > r_2$ and distance $PO_{HW} > r_4$), point P is hidden by none of the water-accessible volumes of the host atoms. Because A_{GO} is a hydrophobic atom ("no" at step h1), this area element is summed up into $S_o(Gc)$ at step i2.

When these steps have been finished for all area elements of A_{GO} ("yes" at step j), all hidden and exposed surface areas of A_{GO} have been calculated. These surface areas are identical to those of the guest molecule in the present case, because the guest molecule has no more atoms to be investigated ("yes" at step k). The above steps are summarized as follows: start \rightarrow a \rightarrow b1 \rightarrow c2 \rightarrow d \rightarrow b2 \rightarrow l \rightarrow m \rightarrow n \rightarrow o \rightarrow p \rightarrow r \rightarrow l \rightarrow m \rightarrow n \rightarrow o \rightarrow p \rightarrow route 1 (q \rightarrow h2 \rightarrow s2 \rightarrow t3) or route 2 (r \rightarrow h1 \rightarrow i2) \rightarrow j \rightarrow k \rightarrow end. At steps t3 and i2 we can get $\Delta S_{ow}(Gc)$ and $S_o(Gc)$, respectively.

Next, we will calculate the exposed and hidden water-accessible areas of the host molecule in the complex shown in Figure 2. Then we must exchange between guest and host at all steps in Figure 3. Let us calculate the water-accessible surface areas of the host hydrophobic atom A_{HO} through the following steps: start \rightarrow a \rightarrow b1 \rightarrow c2 \rightarrow d \rightarrow e \rightarrow f \rightarrow j \rightarrow d \rightarrow e \rightarrow f \rightarrow g \rightarrow b2 \rightarrow l \rightarrow m \rightarrow r \rightarrow h1 \rightarrow i2 \rightarrow j \rightarrow k. At step i2 we get $S_o(Hc)$. The water-accessible surface of this atom intersects that of A_{GO} because eq 4 holds. This atom GO is included in List 2, which enumerates all overlapping guest atoms. Because distance $O_{HO}O_{HW} < r_2 + r_4$ at step c2, A_{HW} is enumerated in List 1. This list includes all overlapping host atoms. At step d, we generate point P on the water-accessible surface of A_{HO} . The world coordinates of point P are expressed by an equation in which GO and r_6 in eq 7 are substituted by HO and r_2 , respectively. At step e we choose A_{HW} enumerated in List 1. In the case where point P is not on arc AB ("yes" at step f, because distance $PO_{HW} < r_4$), the area element of point P does not contribute to any molecular surface area. On the other hand, in the case where point P is on arc AB ("no" at step f, because distance $PO_{HW} > r_4$) and where we have checked all atoms included in List 1 ("yes" at step g), the area element of point P contributes to the water-accessible surface area of the free host molecule, $S(Hf)$. Through steps l–r, we investigate the overlapping of point P with the water-accessible surface of A_{GO} in List 2. Because the present case is "no" at steps m and h1 and "yes" at step r, the area element of point P contributes to $S_o(Hc)$ at

step i2. Steps d–j are repeated for next points on the water-accessible surface of A_{HO} , while step j is “no”.

When all points on A_{HO} have been checked out (“yes” at step j), we will calculate the water-accessible surface areas of A_{HW} (Figure 2) through the following steps: start $\rightarrow a \rightarrow b1 \rightarrow c2 \rightarrow d \rightarrow e \rightarrow f \rightarrow g \rightarrow b2 \rightarrow l \rightarrow m \rightarrow$ route 1 ($r \rightarrow h1 \rightarrow i1$) or route 2 ($n \rightarrow o \rightarrow p \rightarrow q \rightarrow h2 \rightarrow s1 \rightarrow t2$) $\rightarrow j \rightarrow k \rightarrow$ end. In route 1 where step m is “no” (distance $PO_{GO} > r_6$), we can get $Sw(Hc)$ at step i1. In route 2 where step m is “yes” (distance $PO_{GO} < r_6$), we can get $\Delta Swo(Hc)$ at step t2.

For the complex composed of polyatomic host and guest molecules, we can calculate the exposed and hidden water-accessible areas of the complex in the same way with the triatomic complex shown in Figure 2. As the numbers of atoms forming the complex increase, the loops in Figure 3 must be repeated more times.

Experimental Section

First, we investigated six inclusion systems whose crystal structures are available in the literature: α -CyD with methanol²⁸ and propanol²⁹ and β -CyD with ethylene glycol,³⁰ glycerol,³⁰ benzyl alcohol,³¹ and 4-*tert*-butylbenzyl alcohol.³² The three-dimensional structure of complex is expressed in Cartesian coordinate systems with the origin at the center of gravity of a CyD molecule. The CyD molecule is almost symmetric around the x coordinate. The side of its primary hydroxy groups has a negative x value, whereas that of secondary hydroxy groups has a positive x value.

Because no crystal structures of the complexes of α -CyD with hexanol, octanol, and phenol are available in the literature, we assumed that the structure of α -CyD was the same as that of α -CyD in the crystal structure of the α -CyD–propanol complex.²⁹ This crystal structure will be more suitable for complexes of α -CyD than that of an α -CyD hydrate. The atomic coordinates of β -CyD in its complex with 4-methyl-2-pentanol were taken from the crystal structure of a β -CyD hydrate.³³ The structure of γ -CyD was taken from its hydrated structure.³⁴ Because no molecular coordinates of 4-methyl-2-pentanol are available, they were optimized by MM2.

The atomic coordinates of a molecule were determined with, if not specified, standard values of internuclear distances (d), bond angles,³⁵ and atomic radii (r):³⁶ $d_{C-C} = 0.154$ nm, $d_{C-H} = 0.109$ nm, $d_{C-O} = 0.140$ nm, $\angle COC = 109.5^\circ$, $\angle HCH = 109.5^\circ$, $\angle HOC = 109.5^\circ$, $\angle CCC = 109.5^\circ$, $r_H = 0.120$ nm, $r_O = 0.152$ nm, and $r_C = 0.120$ nm. Water radii of $r_w = 0.10$ and 0.14 nm were usually employed for calculations of water-accessible molecular surface areas. Each area element is generated on a water-accessible surface of a molecule and consists of a rectangle with a width of 0.010 nm. A dot was drawn on the center of the element for showing the molecule.

All groups constituting a molecule were classified into either hydrophilic groups or hydrophobic groups. The hydrophilic group includes the hydroxy group and the ether oxygen atom, and the hydrophobic group includes the alkyl group, the phenyl group, and condensed benzene rings.

Calculations and molecular graphics were carried out simultaneously with a personal computer running on Microsoft Windows 95/NT. The relative position of host and guest can be easily varied on the display. All surface areas for a molecule were computed in roughly 25 s.

Results

Stable Structures of CyD Inclusion Complexes. We calculated molecular surface areas of six CyD inclusion systems

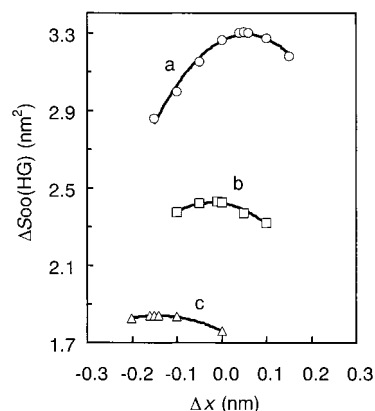


Figure 4. Molecular surface area changes at $r_w = 0.14$ nm, $\Delta S_{oo}(HG)$, with the penetrating depth of guest into the CyD cavity for (a) β -CyD–4-*tert*-butylbenzyl alcohol system, (b) β -CyD–benzyl alcohol system, and (c) α -CyD–propanol system where the penetrating depth is shown as the displacement from their crystal structures.

TABLE 1: Various Water-Accessible Molecular Surface Areas for Three CyD Complexes Whose Structures Have Maximum $\Delta S_{oo}(HG)$ Values

	α -CyD + C_3OH		β -CyD + BzlOH		β -CyD + 4- <i>t</i> -C ₄ -BzlOH	
r_w (nm)	0.10	0.14	0.10	0.14	0.10	0.14
Δx (nm)	−0.20	−0.15	0.02	−0.01	0.00	0.05
$S(HG)$ (nm ²)	9.321	9.887	10.931	11.465	11.342	12.059
$Sw(HG)$ (nm ²)	5.806	6.463	6.793	7.501	7.118	8.061
$So(HG)$ (nm ²)	3.516	3.424	4.138	3.964	4.223	3.998
$\Delta S(HG)$ (nm ²)	2.210	2.693	2.659	3.507	3.165	3.784
$\Delta Sw(HG)$ (nm ²)	0.325	0.427	0.163	0.361	0.105	0.057
$\Delta So(HG)$ (nm ²)	1.885	2.266	2.496	3.147	3.060	3.727
$\Delta S_{sw}(HG)$ (nm ²)	0.203	0.238	0.054	0.149	0.000	0.000
$\Delta S_{so}(HG)$ (nm ²)	0.123	0.186	0.109	0.145	0.105	0.057
$\Delta S_{ow}(HG)$ (nm ²)	0.268	0.427	0.406	0.624	0.332	0.426
$\Delta S_{oo}(HG)$ (nm ²)	1.599	1.839	2.090	2.428	2.729	3.301

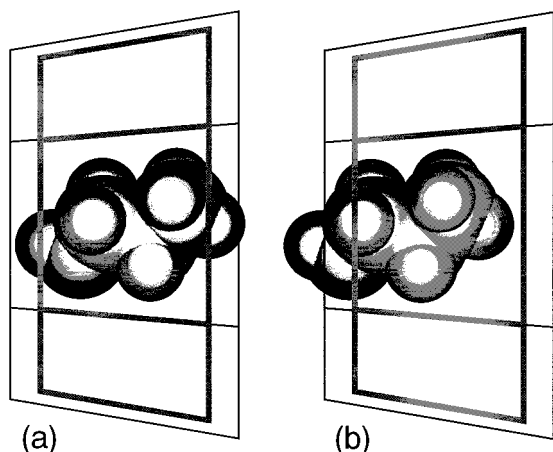
whose crystal structures are available: α -cyclodextrin–methanol,²⁸ α -CyD–propanol,²⁹ β -CyD–ethylene glycol,³⁰ β -CyD–glycerol,³⁰ β -CyD–benzyl alcohol,³¹ and β -CyD–4-*tert*-butylbenzyl alcohol.³² These guests are rather simple aliphatic and aromatic alcohols. The guest molecule was moved along the symmetry axis (x) of CyD. A negative Δx value indicates a displacement of guest from the crystal structure toward the primary hydroxyl side, and a positive value indicates the reverse displacement. We assumed that the structures of host and guest remain unchanged with docking. Although the surface areas at $r_w = 0.10$, 0.14 , and 0.15 nm were calculated actually, the values at 0.10 and 0.14 nm will be reported below.

Figure 4 shows the $\Delta S_{oo}(HG)$ value as a function of Δx for three CyD inclusion systems. Because $\Delta S_{oo}(HG)$ represents the magnitude of contact between the hydrophobic groups of the host and guest in complex, Δx at the maximum of $\Delta S_{oo}(HG)$ will correspond to the stable “solution” structure of the complex. Values of Δx and various surface areas for this stable structure are shown in Table 1. This stable structure is close to the crystal structure, because Δx is very small. When Δx is changed, $So(HG)$ and $\Delta So(HG)$ have a minimum and a maximum, respectively, near $\Delta x = 0$. These areas may be also used for the predictions of stable “solution” structures of complex. The radius of the water molecule used for calculations of water-accessible surface areas slightly influences the estimated solution structure of the complex. The Δx values at $r_w = 0.10$ and 0.14 nm for the stable structures of α -CyD–methanol,²⁸ α -CyD–propanol,²⁹ β -CyD–ethylene glycol,³⁰ β -CyD–glycerol,³⁰ and β -CyD–

TABLE 2: Maximum $\Delta S_{\text{oo}}(\text{HG})$ Values at $r_w = 0.10$ and 0.14 nm and Binding Constants for 11 CyD Inclusion Systems

host	guest	$\Delta S_{\text{oo}}(\text{HG}) (\text{nm}^2)$		$\Delta x (\text{nm})$		$\log K_1$	ref
		0.10	0.14	0.10	0.14		
α -CyD	methanol	0.865	1.046	0.25	0.31	-0.05	37
	propanol	1.599	1.839	-0.20	-0.15	1.37	37
	hexanol	2.393	2.651	<i>a</i>	<i>a</i>	2.94	38
	octanol	2.613	2.962	<i>a</i>	<i>a</i>	3.80	37
	phenol	1.701	1.868	<i>a</i>	<i>a</i>	1.57	39
β -CyD	4-methyl-2-pentanol	1.943	2.170	<i>a</i>	<i>a</i>	2.04	37
	ethylene glycol	0.870	1.012	0.08	0.06	-0.15	40
	glycerol	1.047	1.087	-0.18	-0.05	-0.30	40
γ -CyD	benzyl alcohol	2.083	2.428	0.02	-0.01	2.33	41
	benzene	1.358	2.235	<i>a</i>	<i>a</i>	1.08	42
	naphthalene	2.296	2.805	<i>a</i>	<i>a</i>	2.11	43

^a No crystal structure is available.

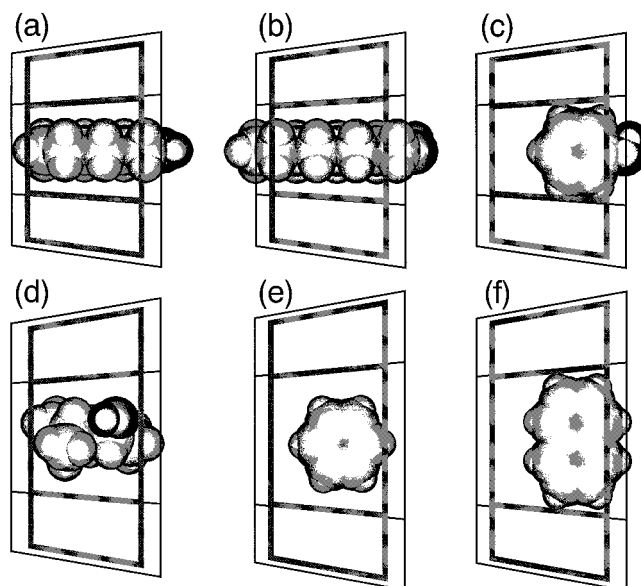
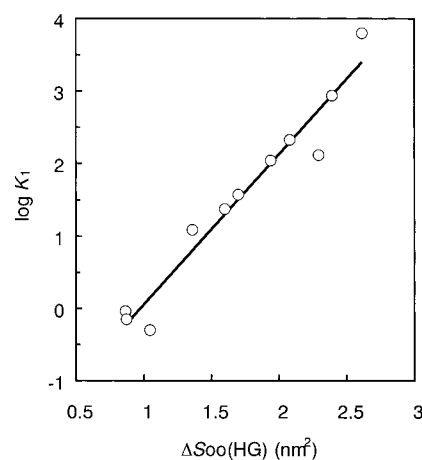
**Figure 5.** (a) Crystal structure and (b) predicted structure of the α -CyD-propanol complex, where a water radius of 0.14 nm is used.

benzyl alcohol³¹ are shown in Table 2. Figure 5 shows the crystal²⁹ and predicted structures of the α -CyD-propanol complex.

No crystal structure of the complex of α -CyD-hexanol is available. On the basis of the crystal structure of the complex of α -CyD-propanol,²⁹ we constructed a structure of the complex of α -CyD-hexanol. That is, we assumed that the structure of α -CyD is identical to that in the complex of α -CyD-propanol. First, propanol in the complex was replaced by hexanol. Then, hexanol was moved along the symmetry axis of α -CyD, and its stable structure was determined at the Δx value exhibiting the maximum $\Delta S_{\text{oo}}(\text{HG})$. The same procedure was applied to the complexes of α -CyD-octanol and α -CyD-phenol. In the case of the β -CyD-4-methyl-2-pentanol system, we used the crystal structure of hydrated β -CyD³³ for β -CyD and the optimized structure of 4-methyl-2-pentanol. This alcohol was docked with β -CyD along its symmetry axis. The hydrated crystal structure of γ -CyD³⁴ was used for complexes of γ -CyD. Figure 6 shows the predicted structures of six complexes whose crystal structures are not available.

Correlation between Binding Constants and $\Delta S_{\text{oo}}(\text{HG})$.

To investigate the correlation between the 1:1 binding constant K_1 and the maximum $\Delta S_{\text{oo}}(\text{HG})$ value, we chose 11 systems rather arbitrarily from the literature.³⁷⁻⁴³ These systems, shown in Table 2, include 5 α -CyDs, 4 β -CyDs, 2 γ -CyDs, and 4 aromatic and 7 aliphatic guests. When two or more values for a binding constant can be found in the literature, such values are generally not in agreement and can differ by as much as several orders of magnitude. For instance, the binding constant

**Figure 6.** Predicted solution structures of complexes of (a) α -CyD-hexanol, (b) α -CyD-octanol, (c) α -CyD-phenol, (d) β -CyD-4-methyl-2-pentanol, (e) γ -CyD-benzene, and (f) γ -CyD-naphthalene, where a water radius of 0.14 nm is used.**Figure 7.** Correlation between the logarithm of the binding constant and $\Delta S_{\text{oo}}(\text{HG})$ at $r_w = 0.10$ nm (\circ) for the 11 host-guest systems shown in Table 2. The line shows eq 20.

for sodium dodecyl sulfate and β -CyD ranges over 3 orders, although reasonable values are recently evaluated and compiled.⁴⁴

In Figure 7, the logarithm of the binding constant is plotted against the $\Delta S_{\text{oo}}(\text{HG})$ value (at $r_w = 0.10$ nm) of the predicted structure for the 11 systems shown in Table 2. The logarithm of the binding constant changes linearly with the maximum $\Delta S_{\text{oo}}(\text{HG})$ value:

$$\log K_1 = 2.074\Delta S_{\text{oo}}(\text{HG}) - 2.016 \quad (R = 0.9741) \quad (20)$$

This correlation becomes worse when we employ a water radius of $r_w = 0.14$ nm:

$$\log K_1 = 1.803\Delta S_{\text{oo}}(\text{HG}) - 2.023 \quad (R = 0.9087) \quad (21)$$

It is notable that Figure 7 includes a variety of guests (monohydroxy, dihydroxy, and trihydroxy alcohols and aliphatic and aromatic compounds) and hosts (α -CyD, β -CyD, and γ -CyD). This result may allow us to predict the binding constant from the calculated $\Delta S_{\text{oo}}(\text{HG})$ value for other CyD inclusion systems.

Discussion

Our calculation and molecular graphics of the water-accessible molecular surface area are based on a dot method. Although the language ASSEMBLER had previously been used,¹⁸ the Windows NT version was developed using Visual C++ in this work. The molecules of host and guest are independently manipulated on the display and all calculated surface areas of their complex are shown. The calculation and molecular visualization usually require about 25 s for the molecules treated in the present work.

The best measure of a tight spatial fit between the guest and host components will be the magnitude of ΔS . The nature of the surface of contact between them will play an important role in their stability. In this work we considered only two kinds of molecular surface, namely, hydrophilic and hydrophobic surfaces. Generally, the contact of surfaces of a similar nature is favorable, whereas the different surfaces will avoid each other. In the present work we focus exclusively on the $\Delta S_{\text{oo}}(\text{HG})$ value. Other ΔS values will play some roles in determining the stable structure and the binding constant, although their contributions were neglected in the present work.

The coefficient of a surface area in the contribution to free energy has the physical meaning of a surface or interfacial tension. The magnitude of the coefficient will depend on the nature of the surface. For instance, the coefficient for aromatic hydrocarbons will be smaller than that for aliphatic hydrocarbons.⁹ This problem must be investigated further, although several sets of such coefficients have been proposed.^{12,25,45,46}

A water radius of 0.14 nm is widely used for proteins,^{10–12} whereas values of 0.10 and 0.15 nm were employed for compounds of low molecular weights.^{9,14,15,18,19} The radius of 0.10 nm corresponds to that of the hydrogen atom. It is not still certain which of these water radii is best consistent with experimental results.

The stable structures of CyD inclusion complexes are proposed on the basis of the maximum $\Delta S_{\text{oo}}(\text{HG})$ values. These structures should be compared to the solution structures. The solution structures of CyD inclusion complexes are not always the same with their crystal structures.⁴⁷ Although the stable position of guest was searched along the symmetry axis of CyD, other probable structures were not investigated. For instance, the rotation of the guest molecule around the symmetry axis of CyD was not taken into consideration, though this effect on $\Delta S_{\text{oo}}(\text{HG})$ will be minor.

We found a linear relationship between $\log K_1$ and $\Delta S_{\text{oo}}(\text{HG})$. As was stated above, we do not know the exact solution structures of the CyD complexes, although the magnitude of $\Delta S_{\text{oo}}(\text{HG})$ does not vary largely with minor changes in structure.

The present approach could be applied to other CyD inclusion systems and to protein–ligand systems. Effective solutions to molecular docking have important implications for molecular recognition, material science, and drug development. A surface-tensions-only model could lower the required computer time enormously, especially for large systems. In fact, driven by practical considerations, surface-tensions-only models have achieved a considerable prominence in the protein-folding literature.¹¹

Nomenclature

Gc	guest in complex
Gf	free guest
Hc	host in complex

Hf	free host
HG	host–guest complex
$S(\text{Gc})$	molecular surface area of complexed guest exposed to water
$S(\text{Gf})$	total molecular surface area of free guest, viz., $S(\text{Gf}) + S_{\text{w}}(\text{Gf})$
$S(\text{Hc})$	molecular surface area of complexed host exposed to water
$S(\text{Hf})$	total molecular surface area of free host, viz., $S(\text{Hf}) + S_{\text{w}}(\text{Hf})$
$S(\text{HG})$	molecular surface area of complex exposed to water and equal to $S(\text{Gc}) + S(\text{Hc})$ and $S(\text{HG}) + S_{\text{w}}(\text{HG})$
$S_{\text{o}}(\text{Gc})$	hydrophobic molecular surface area of complexed guest exposed to water
$S_{\text{o}}(\text{Gf})$	hydrophobic molecular surface area of free guest and equal to $S_{\text{o}}(\text{Gc}) + \Delta S_{\text{o}}(\text{Gc})$
$S_{\text{o}}(\text{Hc})$	hydrophobic molecular surface area of complexed host exposed to water
$S_{\text{o}}(\text{Hf})$	hydrophobic molecular surface area of free host and equal to $S_{\text{o}}(\text{Hc}) + \Delta S_{\text{o}}(\text{Hc})$
$S_{\text{o}}(\text{HG})$	$S_{\text{o}}(\text{Hc}) + S_{\text{o}}(\text{Gc})$
$S_{\text{w}}(\text{HG})$	$S_{\text{w}}(\text{Hc}) + S_{\text{w}}(\text{Gc})$
$S_{\text{w}}(\text{Gc})$	hydrophilic molecular surface area of complexed guest exposed to water
$S_{\text{w}}(\text{Gf})$	hydrophilic molecular surface area of free guest and equal to $S_{\text{w}}(\text{Gc}) + \Delta S_{\text{w}}(\text{Gc})$
$S_{\text{w}}(\text{Hc})$	hydrophilic molecular surface area of complexed host exposed to water
$S_{\text{w}}(\text{Hf})$	hydrophilic molecular surface area of free host and equal to $S_{\text{w}}(\text{Hc}) + \Delta S_{\text{w}}(\text{Hc})$
$\Delta S(\text{Gc})$	molecular surface area of guest hidden from water on complexation, viz., $S(\text{Gf}) - S(\text{Gc})$
$\Delta S(\text{Hc})$	molecular surface area of host hidden from water on complexation, viz., $S(\text{Hf}) - S(\text{Hc})$
$\Delta S(\text{HG})$	decrease in total molecular surface area on complexation, viz., $S(\text{Hf}) + S(\text{Gf}) - S(\text{HG}) = \Delta S(\text{Hc}) + \Delta S(\text{Gc})$
$\Delta S_{\text{o}}(\text{Gc})$	hydrophobic molecular surface area of guest hidden from water on complexation, viz., $S_{\text{o}}(\text{Gf}) - S_{\text{o}}(\text{Gc}) = \Delta S_{\text{o}}(\text{Gc}) + \Delta S_{\text{ow}}(\text{Gc})$
$\Delta S_{\text{o}}(\text{Hc})$	hydrophobic molecular surface area of host hidden from water on complexation, viz., $S_{\text{o}}(\text{Hf}) - S_{\text{o}}(\text{Hc}) = \Delta S_{\text{o}}(\text{Hc}) + \Delta S_{\text{ow}}(\text{Hc})$
$\Delta S_{\text{o}}(\text{HG})$	$\Delta S_{\text{o}}(\text{Hc}) + \Delta S_{\text{o}}(\text{Gc})$
$\Delta S_{\text{w}}(\text{Gc})$	hydrophilic molecular surface area of guest hidden from water on complexation, viz., $S_{\text{w}}(\text{Gf}) - S_{\text{w}}(\text{Gc}) = \Delta S_{\text{wo}}(\text{Gc}) + \Delta S_{\text{ww}}(\text{Gc})$
$\Delta S_{\text{w}}(\text{Hc})$	hydrophilic molecular surface area of host hidden from water on complexation, viz., $S_{\text{w}}(\text{Hf}) - S_{\text{w}}(\text{Hc}) = \Delta S_{\text{wo}}(\text{Hc}) + \Delta S_{\text{ww}}(\text{Hc})$
$\Delta S_{\text{w}}(\text{HG})$	$\Delta S_{\text{w}}(\text{Hc}) + \Delta S_{\text{w}}(\text{Gc})$
$\Delta S_{\text{oo}}(\text{Gc})$	part of guest surface in contact with the hydrophobic group of host in $\Delta S_{\text{o}}(\text{Hc})$
$\Delta S_{\text{oo}}(\text{Hc})$	part of host surface in contact with the hydrophobic group of guest in $\Delta S_{\text{o}}(\text{Gc})$
$\Delta S_{\text{oo}}(\text{HG})$	$\Delta S_{\text{oo}}(\text{Hc}) + \Delta S_{\text{oo}}(\text{Gc})$
$\Delta S_{\text{ow}}(\text{Gc})$	part of guest surface in contact with the hydrophilic group of host in $\Delta S_{\text{o}}(\text{Hc})$
$\Delta S_{\text{ow}}(\text{Hc})$	part of host surface in contact with the hydrophilic group of guest in $\Delta S_{\text{o}}(\text{Gc})$
$\Delta S_{\text{ow}}(\text{HG})$	$\Delta S_{\text{ow}}(\text{Hc}) + \Delta S_{\text{ow}}(\text{Gc})$
$\Delta S_{\text{wo}}(\text{Gc})$	part of guest surface in contact with the hydrophobic group of host in $\Delta S_{\text{w}}(\text{Hc})$
$\Delta S_{\text{wo}}(\text{Hc})$	part of host surface in contact with the hydrophobic group of guest in $\Delta S_{\text{w}}(\text{Gc})$

$\Delta S_{wo}(HG)$	$\Delta S_{wo}(Hc) + \Delta S_{wo}(Gc)$
$\Delta S_{ww}(Gc)$	part of guest surface in contact with the hydrophilic group of host in $\Delta S_{w}(Hc)$
$\Delta S_{ww}(Hc)$	part of host surface in contact with the hydrophilic group of guest in $\Delta S_{w}(Gc)$
$\Delta S_{ww}(HG)$	$\Delta S_{ww}(Hc) + \Delta S_{ww}(Gc)$

References and Notes

- (1) Bender, M. L.; Komiyama, M. *Cyclodextrin Chemistry*; Springer-Verlag: Berlin, 1978; Chapters 2 and 3.
- (2) Szejtli, J. *Cyclodextrin Technology*; Kluwer Academic Publishers: Dordrecht, The Netherlands, 1988; Chapters 2 and 3.
- (3) Saenger, W. In *Inclusion Compounds*; Atwood, J. L., Davies, J. E. D., MacNicol, D. D., Eds.; Oxford University Press: Oxford, U.K., 1984; Vol. 2, Chapter 8.
- (4) Harata, K. In *Inclusion Compounds*; Atwood, J. L., Davies, J. E. D., MacNicol, D. D., Eds.; Oxford University Press: Oxford, U.K., 1991; Vol. 5, Chapter 9.
- (5) Connors, K. A. *Chem. Rev.* **1997**, 97, 1325.
- (6) Sherrod, M. In *Spectroscopic and Computational Studies of Supramolecular Systems*; Davies, J. E. D., Ed.; Kluwer Academic Press: Dordrecht, The Netherlands, 1992; Chapter 7.
- (7) Connors, K. A. *J. Pharm. Sci.* **1996**, 85, 796.
- (8) Mayer, B.; Marconi, G.; Klein, C.; Koeler, G.; Wolschann, P. *J. Inclusion Phenom. Mol. Recognit. Chem.* **1997**, 29, 79.
- (9) Hermann, R. B. *J. Phys. Chem.* **1972**, 76, 2754.
- (10) Honig, B.; Sharp, K.; Yang, A.-S. *J. Phys. Chem.* **1993**, 97, 1101.
- (11) Eisenberg, D.; McLachlan, A. D. *Nature* **1986**, 319, 199.
- (12) Hawkins, G. D.; Cramer, C. J.; Truhlar, D. G. *J. Phys. Chem.* **1997**, 101, 7147.
- (13) Pearlman, R. S. In *Physical Chemical Properties of Drugs*; Yalkowsky, S. H., Sinkula, A. A., Valvani, S. C., Eds.; Marcel Dekker: New York, 1980; Chapter 10.
- (14) Funasaki, N.; Hada, S.; Neya, S.; Machida, K. *J. Phys. Chem.* **1984**, 88, 5786.
- (15) Funasaki, N.; Hada, S.; Neya, S. *J. Phys. Chem.* **1985**, 89, 3046.
- (16) Tanford, C. *The Hydrophobic Effect: Formation of Micelles and Biological Membranes*, 2nd ed.; John Wiley and Sons: New York, 1980; Chapter 6.
- (17) Funasaki, N.; Shim, H.-S.; Hada, S. *J. Phys. Chem.* **1992**, 96, 2754.
- (18) Ishikawa, S.; Hada, S.; Funasaki, N. *J. Phys. Chem.* **1995**, 99, 11508 and references therein.
- (19) Funasaki, N.; Hada, S.; Neya, S. *J. Chromatogr.* **1986**, 361, 33.
- (20) Funasaki, N.; Hada, S.; Neya, S. *Anal. Chem.* **1993**, 65, 1861.
- (21) Funasaki, N.; Hada, S.; Neya, S. *J. Med. Chem.* **1983**, 26, 686.
- (22) Serrano, L.; Neira, J.-L.; Sancho, J.; Fersht, A. R. *Nature* **1992**, 356, 453.
- (23) Doucet, J.-P.; Weber, J. *Computer-Aided Molecular Design: Theory and Applications*; Academic Press: London, 1996; Chapters 8 and 13.
- (24) Searle, M. S.; Williams, D. H.; Gerhard, U. *J. Am. Chem. Soc.* **1992**, 114, 10697.
- (25) Ajay; Murcko, M. A. *J. Med. Chem.* **1995**, 38, 4953.
- (26) Ooi, T.; Oobatake, M.; Neméthy, G.; Scheraga, H. D. *Proc. Natl. Acad. Sci. U.S.A.* **1987**, 84, 3086.
- (27) Ishikawa, S.; Neya, S.; Funasaki, N. *J. Phys. Chem. B* **1998**, 102, 2502.
- (28) Hingerty, B.; Saenger, W. *J. Am. Chem. Soc.* **1976**, 98, 3357.
- (29) Saenger, W.; McMullan, R. K.; Fayos, J.; Mootz, D. *Acta Crystallogr.* **1974**, B30, 2019.
- (30) Gessler, K.; Steiner, T.; Koellner, G.; Saenger, W. *Carbohydr. Res.* **1993**, 249, 327.
- (31) Hirata, K.; Uekama, K.; Otagiri, M.; Hirayama, F.; Ohtani, Y. *Bull. Chem. Soc. Jpn.* **1985**, 58, 1234.
- (32) Mentzafos, D.; Mavridis, I. M.; Tsoucaris, G. G. *Acta Crystallogr.* **1991**, B47, 746.
- (33) Lindner, K.; Saenger, W. *Carbohydr. Res.* **1982**, 99, 103.
- (34) Harata, K. *Bull. Chem. Soc. Jpn.* **1987**, 60, 2763.
- (35) Clark, T. *A Handbook of Computational Chemistry*; John Wiley and Sons: New York, 1985; Appendix C.
- (36) Bondi, A. *J. Phys. Chem.* **1964**, 68, 441.
- (37) Matsui, Y.; Mochida, K. *Bull. Chem. Soc. Jpn.* **1979**, 52, 2808.
- (38) Rekharsky, M. V.; Schwartz, F. P.; Tewari, Y.; Goldberg, R. N.; Tanaka, M.; Yamashoji, Y. *J. Phys. Chem.* **1994**, 98, 4098.
- (39) Bertrand, G. L.; Faulkner, J. R., Jr.; Han, S. M.; Armstrong, D. W. *J. Phys. Chem.* **1989**, 93, 6863.
- (40) Buvari, A.; Szejtli, J.; Barcza, L. *J. Inclusion Phenom.* **1983**, 1, 151.
- (41) Rymden, M.; Carlfors, J.; Stilbs, P. *J. Inclusion Phenom.* **1983**, 1, 159.
- (42) Tucker, E. E.; Christian, S. D. *J. Am. Chem. Soc.* **1984**, 106, 1942.
- (43) Fujiki, M.; Deguchi, T.; Sanemasa, I. *Bull. Chem. Soc. Jpn.* **1988**, 61, 1163.
- (44) Funasaki, N.; Yodo, H.; Hada, S.; Neya, S. *Bull. Chem. Soc. Jpn.* **1992**, 65, 1323 and references therein.
- (45) Wesson, L.; Eisenberg, D. *Protein Sci.* **1992**, 1, 227.
- (46) Sitkoff, D.; Sharp, K. A.; Honig, B. *J. Phys. Chem.* **1994**, 98, 1978.
- (47) Alderfer, J. L.; Eliseev, A. V. *J. Org. Chem.* **1997**, 62, 8225.