Hydrophobic Hydration Processes. I: Dual-Structure Partition Function for Biphasic Aqueous Systems

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ABSTRACT: The thermodynamic properties of hydrophobic hydration processes have been analyzed and assessed. The thermodynamic binding functions result to be related to each other by the mathematical relationships of an ergodic algorithmic model (EAM). The active dilution \( d_A \) of species \( A \) in solution is expressed as \( d_A = 1/(\Phi - x_A) \) with thermal factor \( \Phi = T - (C_P A/R) \) and \( 1/x_A = d_{id(A)} \), where \( d_{id(A)} \) is ideal dilution. Entropy function is set as \( S = f(d_{id(A)}, T) \). Thermal change of entropy (i.e., entropy intensity change) is represented by the equation \((dS)_{th} = C_p \ln T\). Configuration change of entropy (i.e., entropy density change) is represented by the equation \((dS)_{dual} = (R \ln x_A - T)\). Because every logarithmic function in thermodynamic space corresponds to an exponential function in probability space, the sum functions \( \Delta H_{dual} = (\Delta H_{mot} + \Delta H_{id}) \) and \( \Delta S_{dual} = (\Delta S_{mot} + \Delta S_{id}) \) of the thermodynamic space give birth, in exponential probability space, to a dual-structure partition function \( \{DS-PF\} \): \( \exp(-\Delta G_{dual}/RT) = K_{dual} = (K_{mot} K_{id}) = \{exp\left(-\Delta H_{dual}/RT\right)\} \cdot \{exp\left(-\Delta H_{id}/RT\right)\} \cdot \{exp\left(-\Delta S_{mot}/R\right)\} \cdot \{exp\left(-\Delta S_{id}/R\right)\} \). Every hydrophobic hydration process can be represented by \( \{DS-PF\} = \{M-PF\} \cdot \{T-PF\} \), indicating biphasic systems. \( \{M-PF\} = f(T, d_{id(A)}) \), concerning the solute, is monocentric and produces changes of entropy density, contributing to free energy \( -\Delta G_{mot} \) whereas \( \{T-PF\} = g(T) \), concerning the solvent, produces changes of entropy intensity, not contributing to free energy. Entropy density and entropy intensity are equivalent and summed with each other (i.e., they are ergodic). From the dual-structure partition function \( \{DS-PF\} \), the ergodic algorithmic model (EAM) can be developed. The model EAM consists of a set of mathematical relationships, generating parabolic convoluted binding functions \( R \ln K_{dual} = -\Delta G_{dual}/T = f(1/T)^* g(T) \) and \( RT \ln K_{dual} = -\Delta G_{dual} = f(T)^* g(ln T) \). The first function in each convoluted couple \( f(1/T) \) or \( f(T) \) is generated by \( \{M-PF\} \), whereas the second function, \( g(T) \) or \( g(ln T) \), respectively, is generated by \( \{T-PF\} \). The mathematical properties of the thermodynamic functions of hydrophobic hydration processes, experimentally determined, correspond to the geometrical properties of parabolas, with constant curvature amplitude \( C_{amp} = 0.7071/\Delta C_{p,hid} \). The dual structure of the partition function conforms to the biphasic composition of every hydrophobic hydration solution, consisting of a diluted solution, with solvent in excess at constant potential.

1. INTRODUCTION

In 1980, Lumry\(^1\) proposed to consider the thermodynamic functions enthalpy and entropy in biochemical reactions as divided into two parts, namely, thermal (or compensative) and motive (or work) functions, respectively. Moreover, Lumry regretted that very rarely the dual nature of water had been considered in biochemical equilibria. Lumry\(^2\) observed that enthalpy, entropy, and volume data obtained for processes studied in aqueous solvent generally have been assumed to apply to a solute process, without consideration of the coupling between the process and the two-state equilibrium of water. It followed then that significant chemistry knowledge deduced from reactions in aqueous media may be wrong. Most free-energy information is likely to remain uncomplicated, but enthalpy, internal energy, entropy, and volume data are generally suspected, since have they been rarely analyzed as to take the two species of water into account. Another point touched by Lumry, again about biochemical equilibria in aqueous solvents, concerned the peculiar properties of the thermodynamic functions enthalpy and entropy. Lumry recommended to consider enthalpy and entropy as composed of two parts each, thermal and motive. Lumry,\(^3\) however, stated that thermal and motive parts of enthalpy and entropy were not usually experimentally determinable. Lumry much appreciated also the property observed by Benzinger,\(^3\) supported by Lee and Graziano,\(^4\) that some of the components of the thermodynamic functions compensate each other and do not contribute to free energy.

We have been studying\(^5\) for the last 10 years the thermodynamic properties of the hydrophobic hydration processes, including the reactions of protein denaturation and micelle formation, specifically considered by Lumry. In contrast, however, with the pessimistic opinion expressed by Lumry, we have succeeded to calculate numerically thermal and motive components of enthalpy and entropy in every one of the many hydrophobic processes examined, by taking...
advantage of constant hydrophobic heat capacity \( \Delta C_{\text{ph}} \). By assuming that water is composed of cluster \( W_\ell \) and iceberg \( W_{\text{ice}} \), we have built up a molecular model for hydrophobic hydration processes that yields very significant self-consistent results, notwithstanding that the experimental data are referred to apparently different processes, from protonation of carboxylato anions and noble gas solubility in water to protein denaturation, micelle formation, and many others. Several points will be addressed:

(i) Dual-structure partition function \( \{ \text{DS-PF} \} = \{ \text{M-PF} \} \) is indicative of the biphasic structure of every hydrophobic hydration system, with the motive function \( \{ \text{M-PF} \} = f(T, d_{\text{dil}}/A) \) referred to the reacting solute and the thermal function \( \{ \text{T-PF} \} = g(T) \) referred to the excess solvent, respectively.

(ii) Dual-structure partition function \( \{ \text{DS-PF} \} \) is valid for every hydrophobic hydration system.

(iii) Hydrophobic heat capacity \( \Delta C_{\text{ph}} \) is constant in each compound, independent of \( T \).

(iv) Thermal free energy \( \langle \Delta G_{\text{dil}}/T \rangle \) referred to \( \{ \text{T-PF} \} \) is invariably zero.

(v) The second law of thermodynamics is related to both entropy intensity and entropy density.

(vi) Every dual-structure partition function of probability space generates, in thermodynamic space, parabolic convoluted binding functions \( R \ln K_{\text{dil}} = \langle \Delta G_{\text{dil}}/T \rangle = \langle f(T)g(T) \rangle \) and \( RT \ln K_{\text{dil}} = \langle \Delta G_{\text{dil}} \rangle = \langle f(T)g(T) \rangle \), constituting an ergodic algorithmic model (EAM). In consequence, constant hydrophobic heat capacity \( \Delta C_{\text{ph}} \) is inversely proportional to the geometrical constant curvature amplitude \( C_{\text{ampl}} = 0.7071/\Delta C_{\text{ph}} \) of both parabolic binding functions, results to be an invariable property of every hydrophobic hydration process.

(vii) Condition of null thermal free energy \( \langle \Delta G_{\text{dil}}/T = 0 \rangle \) is an invariable property of the thermal partition function \( \{ \text{T-PF} \} \), which is referred to a large statistical molecule population (solvent) and can be treated by methods of statistical thermodynamics.

(viii) Motive partition function \( \{ \text{M-PF} \} \) is monocentric, with linear van’t Hoff equation \( \langle \Delta G_{\text{mot}}/T \rangle = f(1/T) \) in thermodynamic space, as in any normal monophasic system with constant \( K_{\text{mot}} \). The partition function \( \{ \text{M-PF} \} \), being referred to an ensemble composed by very few elements (mols, solute) ruled by binomial distribution, cannot be treated by methods of statistical thermodynamics.

(ix) Consistency or inconsistence of ergodic algorithmic model (EAM) with computer simulations of free-energy functions is discussed. Computer simulations concerning the dual-structure partition function \( \{ \text{DS-PF} \} \) requires necessarily the introduction of quasi-chemical approximations,\(^9\) associating molecule statistical distributions of \( \{ \text{T-PF} \} \) to mole binomial distribution of \( \{ \text{M-PF} \} \). The potential distribution theorem (PDT),\(^7\) which is referred to a nonexistent monophasic system, is inconsistent with the ergodic algorithmic model (EAM).

(x) The essential reaction of the hydrophobic effect is, in class A, the reaction of iceberg formation with phase transition

\[
A\{ \xi_w, W_l(\text{solvent}) \rightarrow \xi_w, W_{\text{ice}}(\text{iceberg}) \}
\]

whereas in class B, the reaction, with opposite phase transition, is

\[
B\{ -\xi_w - \xi_w, W_{\text{ice}}(\text{iceberg}) \rightarrow \xi_w, W_l(\text{solvent}) \}
\]

with iceberg reduction. It is worth mentioning that the processes of iceberg formation in class A and iceberg reduction in class B, taking place in the solvent, with reduction or increment of solvent volume, respectively, produce changes in the thermodynamic properties of the solute.

This manuscript is the first part of the three-part study of hydrophobic hydration processes:

I. Dual-structure partition function for biphasic aqueous systems.

II. Entropy density and entropy intensity: ergodic algorithmic model (EAM).

III. Validation of (EAM) model.

In Part II, the molecular interpretation of the entropy intensity changes and of the entropy density changes is discussed. The variability of entropy density is bound, at constant temperature \( T \), to changes of ideal dilution \( d_{\text{dil}}/A = 1/x_4 \) multiplied by the thermal factor \( f(r_{\text{m}}) = T^{-(r_{\text{m}}/8)} \). The thermal factor represents, at constant temperature, the thermal energy associated with each molecule. \( r_{\text{m}} \) represents the squared mean sojourn time of each molecule. Sojourn time is the time spent by each molecule to run 1 length unit. The variability of entropy intensity is bound to changes of velocity of the molecules, produced by changes of temperature, through the same thermal factor, in systems, like as a pure liquid, whereby no concentration change is possible. Both processes, variation of entropy density and variation of entropy intensity, produce changes of energy dispersive, i.e., changes of entropy. The connections of EAM with computer simulations will be discussed. Computer simulations will be conditioned by the molar reactions taking place in every hydrophobic hydration process.

In Part III, the analysis is extended to a large population of compounds of very different size and very different molecular structure. The statistical analysis over a population of about 80 compounds with about 600 experimental points has confirmed that EAM is valid in every hydrophobic hydration process, leading to unitary values of entropy change and enthalpy change, with variability at the limit of the experimental error. Statistical validation states the general applicability of EAM and indicates that the same properties of hydrophobic hydration process will be found in every such process taking place in biological ambient and in every process of drug design.

2. RESULTS AND DISCUSSION

2.1. Dilution, Thermal Energy Dispersal, and Entropy.

For statistical thermodynamics, the number of possible locations (configurations) for a solute molecule in the whole solution volume is a measure of the state probability of that solute. The larger the number of possible locations, the larger the solvent volume. The state probability of a solute, therefore, is proportional to the volume in which the solute is dissolved, i.e., to solute dilution. The identification of solvent-to-solute ratio (dilution) as homologous with configuration multiplicity is an important connection to statistical thermodynamics. A basic statistical setting is the relationship between state multiplicity, state probability, and entropy density. If we suppose to subdivide the solvent into many submicroscopic cells, we can consider each cell as a possible location for a
solute molecule. The larger the number of accessible cells, the more probable a molecule can find a cell for location. The cell multiplicity, therefore (expressed as the number of solvent molecules per solute unit), identifies the statistical state probability \( \Omega \), calculated by Boltzmann equation

\[
\Omega = \exp(S/k_B)
\]

In a solution, the number of available cells is directly proportional to dilution (expressed as solvent-to-solute ratio): consequently, dilution, which is a number expressing the dispersion of solute molecules, is also homologous with the number expressing statistical state probability \( \Omega \). If the concentration \( c_A \) is expressed in molar fraction (solute-to-solvent ratio), its reciprocal, ideal calculated dilution \( d_{\text{id}}(A) = 1/x_A \), represents the solvent-to-solute ratio. Ideal dilution \( d_{\text{id}}(A) \), therefore, which is a parameter of matter dispersion, corresponds to an exponential probability factor in probability space

\[
d_{\text{id}}(A) = \exp(S/R)
\]

whereby we show how ideal dilution \( d_{\text{id}}(A) \) is an exponential function homologous with statistical state probability \( \Omega \) of statistical thermodynamics of eq 1.

The molecules, however, tend to disperse over all of the available accessible microcells, stirred by thermal energy in such a way that the molecules carry thermal energy to every available accessible microcell. This point has been stressed by Lambert\(^{10}\) who has launched a campaign to inform students that using the simple numerical probability without any mention of energetic involvement might be misleading. Lambert speaks of energetic "enablement". The energetic involvement recommended by Lambert is essential to obtain any entropy change from matter dispersion change.

Regarding this energetic involvement, we can recall an analogy. If we let a drop of purple solute fall into a colorless liquid solvent, the thermal energy will bring the solute molecules to disperse all over the whole volume of the solvent, thus obtaining a pale pink color. Suppose that energy identifies with a purple layer covering each molecule: the color is dispersed all over the solvent volume. Dispersion of energy is analogous to dispersion of color. With dispersion of energy measured by the thermodynamic function entropy density \( S_{\text{dens}} \), we can suppose \( S_{\text{dens}} \) to be inversely proportional to the intensity of the solution color. For the moving solute molecules, every microcell of solvent is potentially a location. Each solute molecule, however, carries with it a portion of thermal energy, supposed to be purple, in such a way that dispersion of matter measured by ideal dilution \( d_{\text{id}}(A) = 1/x_A \) becomes at the same time dispersion of "purple red" energy, i.e., entropy density. Without thermal agitation, the purple solute molecules would have been resting at the initial point, so the same resting would have occurred for energy, and as a consequence, we could not observe any entropy density change. The dispersion of matter \( d_{\text{id}}(A) \), therefore, is proportional to a change of entropy density in the thermodynamic space: we can write, at constant temperature, the entropy density differential

\[
(dS_{\text{dens}})/T = R \ln d_{\text{id}}(A)
\]

We can introduce the energetic "enablement" recommended by Lambert by substituting the concentration \( x_A \) by activity \( a_A \), where \( a_A = \Phi \cdot x_A \). The Lambert’s thermal energy factor (THEF, with \( \Phi = T^{(C_{\text{rho}}/R)} \) representing the energetic involvement recommended by Lambert’s is a measure of the thermal energy, supposed to be "purple red", associated with each molecule. THEF is a source of ergodicity of the chemical systems. The ideal dilution \( d_{\text{id}}(A) = 1/x_A \) is transformed into the active dilution \( d_A = 1/a_A \): the reciprocal factor \( (1/\Phi) \), therefore, transforms the parameter of matter dispersion \( d_{\text{id}}(A) = 1/x_A \) into the parameter of energy dispersion \( d_A = 1/a_A \) and hence into an entropy parameter. We can consider that the entropy \( S_{\text{dens}} \) can be expressed as the function of activity of \( A \) as

\[
dS = -R \ln a_A = R \ln d_A
\]

The active dilution \( d_A \) can be set

\[
d_A = (1/x_A) \cdot (1/\Phi) = (1/x_A) \cdot (T^{(C_{\text{rho}}/R)}) = d_{\text{id}}(A)
\]

and the differential of entropy can be rewritten as

\[
dS = Rd\ln d_A = R \ln \{(1/x_A) \cdot (1/\Phi)\}
\]

By developing eq 6, we obtain

\[
dS = R \ln d_A = R \ln d_{\text{id}}(A) + dS_{\text{dens}}
\]

\[
= R(\ln d_{\text{id}}(A))/T + (C_{\text{p},A} \ln T) d\ln d_A
\]

\[
= R \ln d_{\text{id}}(A) + (C_{\text{p},A})dT/T
\]

\[
= R \ln d_{\text{id}}(A) + \delta_{\text{rev}}Q_{p}/T
\]

The two terms of eq 7, representing entropy density and entropy intensity differential changes, respectively, are related to the changes of energy dispersion at molecular level: the first term indicates the change of energy dispersion in space, i.e., change of entropy density, whereas the second term corresponds to the change of energy dispersion in time, i.e., change of entropy intensity, respectively (see Part II, Section 2, for molecular interpretations of entropy density and entropy intensity).

Equation 7 implies the formulation of the entropy function, bound to an extended second law of thermodynamics (see Appendix A). The reappraisal of the traditional formulation of the second law is necessary because the usual expression of the second law, with condition to entropy, \( dS = \delta_{\text{rev}}Q_{p}/T \geq 0 \) (being \( \delta_{\text{rev}}Q_{p}/T = C_p dT/T = C_p \ln T \geq 0 \)), is inadequate. It is, in fact, clearly referred to the only thermal entropy change, or entropy intensity change due to heat transfer, in conformity with Clausius’ definition of the second law. The existence of the configuration entropy changes or entropy density changes requires the extension of the validity of the second law. Entropy intensity and entropy density are equivalent (ergodic) and are summed up or subtracted to each other.

At constant temperature (\( \ln T = 0 \), the entropy change reduces to eq 3, but we have to remind the reader that the reciprocal \( \text{THEF} (1/\Phi \neq 1) \) is implicitly active, even if constant for \( [\delta(1/\Phi)/\delta T]/T = 0 \). In such isothermal conditions, the variation of entropy density as a function of ideal dilution \( \ln d_{\text{id}}(A) = \ln (1/x_A) \) can be measured by experimental determinations of variations of \( x_A \): every potentiometer or pH meter, in fact, becomes, at constant temperature, an effective entropy density meter. Entropy,
therefore, is a function of ideal calculated dilution $d_{d(A)}$, which is homologous, at constant temperature, with state probability.

We can now search for other functions homologous with state probability and dependent on dilution. To endeavor these functions, we recall the Boltzmann equation for statistical ensemble

$$S = k_B \ln \Omega$$

where $\Omega$ is a partition function referred to an ensemble of molecules as the function of $k_B = 1.3806 \times 10^{-23}$ J K$^{-1}$. The number $\Omega$ is an extremely large quantity calculated by statistical mechanics methods and not accessible by experiment. We calculate the Nth root of $\Omega$ ($N$ is Avogadro number: $N_A = 6.022 \times 10^{23}$) and transform $\Omega$ into the partition function $Z_M$ referred to a population of moles as the function of $R$ ($R = 8.31451$ J K$^{-1}$ mol$^{-1}$)

$$S = (N_A k_B) \ln \left(1/N_0\right) = R \ln Z_M$$

where $Z_M$ is a molar partition function. $Z_M$ is, in principle, experimentally accessible, being on mole chemical scale. By differentiation, we obtain

$$(dS)_T = R \ln Z_M = R \ln d_{d(A)}$$

at constant $T$, whereby we put in evidence, in comparison to eq 3, the parallelism between entropy density, logarithm of partition function, and logarithm of ideal dilution.

At the same time, however, this is the point strongly supported by Lambert: dilution is a measure of energy dispersion (i.e., energy dispersion (or energy dilution) means entropy concentration) because the molecules, moved by thermal energy, represented by the factor $(1/\Phi)$ (reciprocal THEF) in eq 5, tend to spread over every accessible solvent cell, thus changing dilution. This process goes on until it reaches the minimum concentration and consequently the maximum dilution compatible with the system conditions. In such a way, dilution from parameter of matter dispersion becomes a parameter of energy dispersion, i.e., a parameter of entropy density. Lambert regrets that statistical thermodynamic authors insist on the probabilistic aspect of multiplicity without any mention of energy involvement. We have shown above how the energy "enablement" of configuration entropy, as suggested by Lambert, can be explained by introducing the reciprocal of energetic factor THEF $(1/\Phi = T^{G(x)/R})$, multiplying the ideal calculated dilution $d_{d(A)}$. In contrast, pure thermal changes of entropy intensity can be observed whenever heat dispersion in time, due to changed velocity of the molecules, takes place even without any concentration change $(1/x_A = 1/\ln N_0)$, as for example in a solvent or in a pure nonreacting liquid. For the solvent or a pure liquid, in fact, it is constitutionally impossible to define a concentration change. In these systems, therefore, we can observe changes of entropy intensity only.

2.2. Configuration Change of Entropy: Entropy Density. Being on search for thermodynamic functions depending on dilution, which is homologous with configuration, we have analyzed the formation constant of the equilibrium $A + B = AB$

$$K = x_{AB}/(x_B x_A)$$

By considering that $d_{d(L)} = 1/x_A$, $d_{d(B)} = 1/x_B$, and $d_{d(AB)} = 1/c_{AB}$, the formation constant can be rewritten as a ratio of ideal dilutions

$$K = (d_{d(L)} \cdot d_{d(B)})/d_{d(AB)}$$

with clear connection to the settings of statistical thermodynamics, whereby the configurations as state probability are homologous with ideal calculated dilutions. It is worth noting, in fact, that if the concentration is expressed in molar fraction, the ideal dilution $d_{d(L)} = 1/x_A$ represents the molar (solvent-to-solute) ratio. The dilution is homologous with the statistical partition function $\Omega$, which represents the molecular (solvent-to-solute) ratio. By calculating the logarithm $R \ln K$ at temperature $T$, we move from probability space ($K$) to thermodynamic space

$$(R \ln K)_T = +R \ln d_{d(L)} + R \ln d_{d(B)} - R \ln d_{d(AB)}$$

and by recalling that for component $A$

$$\Delta S_A = R \int_{1}^{d_{d(A)}} d\ln d_{d(A)}$$

and so on for other terms, we can write

$$(R \ln K)_T = +\Delta S_A + \Delta S_B - \Delta S_{AB} = \Delta S_{tot}$$

showing that $(R \ln K)_T$, being formed by a sum of entropy density terms, is itself an entropic function. The character of total entropy density of $R \ln K = (-\Delta G°/T)$ conforms to the statement of statistical thermodynamics that, according to the second law of thermodynamics, a system assumes the configuration of maximum entropy, at thermodynamic equilibrium. This state probability, in fact, maximizes the discrete Gibbs entropy.

In general terms, $\ln K$ is a specific value of a general equilibrium quotient $Q_K$. By considering that $R \ln d_{d(L)} \sim R \ln d_{d(AB)}$, the differential of equilibrium quotient $Q_K$ can be expressed as

$$R \ln Q_K = R \ln d_{d(A)}$$

A change of $Q_K$ corresponds, therefore, at constant temperature, to a change of dilution and hence to a change of entropy density. The partition function in probability space is homologous with dilution. The logarithm of equilibrium quotient, the logarithm of the partition function, and the logarithm of equilibrium constant can be reported along the dilution axis in the diagram (Figure 1), where we report the

![Figure 1](image-url)
vector representation of Gibbs equation in thermodynamic space. In this diagram, we report the configuration (dilution) change of entropy (entropy density) on the abscissa axis and the thermal change of entropy (entropy intensity) on the ordinate axis.

The next function analyzed has been the chemical potential (partial molar function)

\[
\left(\frac{\partial G}{\partial n_j}\right)_{P,T,x_j} = \mu_j
\]

(17)
a function introduced to represent the dependence of free energy from concentration of each reactant

\[
\mu_A^{\text{c}} = \mu_A^{\text{c},0} + RT \ln x_A
\]

(18)
with \(x_A\) the concentration of \(A\), in molar fraction.

For a given temperature, a molecule has a higher chemical potential in a high concentration sector and a lower chemical potential in a low concentration sector.

\[
\left(\frac{\mu_j}{T}\right) = \left(\frac{\mu_j^{\text{c},0}}{T}\right) + R \ln x_A
\]

(19)
after differentiation

\[
d\left(\frac{\mu_j}{T}\right) = \frac{\partial}{\partial x_A} \left[\mu_j^{\text{c},0} + RT \ln x_A\right] = \left(\frac{d\mu_j}{T}\right)_T
\]

(20)
and with sign changed, we obtain

\[
d\left(-\frac{\mu_j}{T}\right) = -R \ln x_A = R \ln d_{\text{d}(A)} = \left(\frac{dS_{\text{den}}}{T}\right)_T
\]

(21)
This equation shows how the differential \(d(-\mu_j/T)\) also is a configuration change of entropy density and can be reported on the dilution axis of the diagram in Figure 1.

The Gibbs equation, referred to a monocentric partition function, is represented in Figure 1 as a vector (bold type) composition

\[
-\Delta G^{\text{c},0}/T = -\Delta H^{\text{c},0}/T + \Delta S^{\text{c},0}
\]

(22)
The reaction is assumed to be exothermic (\(\Delta H^{\text{c},0} < 0\)). We note that \(\Delta G^{\text{c},0}/T\) (thermal entropy intensity) is by construction necessarily equal to \(\Delta S_{\text{H}}\) (change of entropy density)

\[
-\Delta H^{\text{c},0}/T = \Delta S_{\text{H}}
\]

(23)
Both are, in fact, legs of an isosceles right triangle. The equivalence demonstrates the ergodic property of the thermodynamic system. In fact, eq 23 states that an enthalpy divided by temperature \(T\) (i.e., entropy intensity) is transformed by projection onto the dilution axis into a configuration change of entropy (i.e., entropy density). Thus, we find on the \(x\) axis a total change of entropy density vector \(\Delta S_{\text{TOT}}^{\text{c},0} = -\Delta G^{\text{c},0}/T\), which represents the sum of the entropy vectors \(\Delta S_{\text{H}}\) (entropy density equivalent to entropy intensity) and \(\Delta S^{\text{c},0}\) (change of entropy density)

\[
\Delta S_{\text{H}} + \Delta S^{\text{c},0} = \Delta S_{\text{TOT}}^{\text{c},0}
\]

(24)
This vector can also be represented along dilution axis as entropy density function (\(dS_T\) \(\equiv dS_{\text{den}}\)). Every configuration change of entropy or change of entropy density in reacting mole ensembles (REMEs)\(^b\) can be reported, as dilution-equivalent on \(x\) axis (Table 1). We want, however, to stress once again the point that the dilution differential \(R \ln d_{\text{A}}\) is actually enabled to represent dispersion of energy, as a change of entropy density, only because the ideal calculated dilution \(d_{\text{d}(A)}\) is associated with the active dilution \(d_{\text{A}}\) to the reciprocal thermal factor THEF (1/\(\Phi = T^{G_{\text{ss}}(1)}\)) (cf. eq 5).

Table 1. Dilution-Equivalent Entropy Density Functions, in REME Ensembles

| Function | Expression |
|----------|------------|
| \(dS\) | \(=-R \ln x_A = R \ln d_{\text{d}(A)}\) \(dS_T = R \ln d_{\text{d}(A)}\) |
| \(dS\) | \(=-R \ln x_A = R \ln d_{\text{A}}\) \(dS_T = R \ln d_{\text{A}}\) |
| \(dS\) | \(=R \ln Q_k = R \ln d_{\text{A}}\) \(dS_T = nR \ln d_{\text{d}(A)}\) |
| \(dS\) | \(=-\Delta G/T = R \ln d_{\text{A}}\) \(dS_T = nR \ln d_{\text{d}(A)}\) |
| \(dS\) | \(=-\Delta G/T = R \ln d_{\text{A}}\) \(dS_T = nR \ln d_{\text{d}(A)}\) |

One special point is worth noting, concerning the diagram in Figure 1 and van’t Hoff function. By determining the equilibrium constant \(R \ln K\) at different temperatures for any kind of reaction, we measure in any case changes of entropy density, which can be reported on dilution axis. Then, by calculating the derivative \(\partial (R \ln K)/\partial (1/T)\) for these configuration data (van’t Hoff equation), we “calculate” the molar enthalpy \(\Delta H^{\text{c},0}\). In a general regular reaction, the derivative \(\partial (R \ln K)/\partial (1/T)\) is a constant \(\Delta H^{\text{c},0}\). In hydrophobic reactions, we obtain the experimental enthalpy \(\Delta H_{\text{ideal}}\) that varies following a straight line as in eq 31. In any case, \(\Delta H/T\) is an entropy intensity: this means that, by applying van’t Hoff equation, we calculate entropy intensity changes from measurements of entropy density. If, in a different experiment on the same reaction, we employ a calorimeter to measure reaction enthalpy and obtain an experimental value of molar enthalpy \(\Delta H^{\text{c},0}\) or \(\Delta H_{\text{ideal}}\) equal to that previously “calculated” from concentration (i.e., configuration) determinations (cf. eq 23). This result is possible because of the ergodic properties of the thermodynamic systems (cf. Part II, Section 2\(^{11}\)). The ergodic condition defines the equivalence between energy dispersion in time, or entropy intensity, due to temperature \(T\) and hence velocity of molecules, and energy dispersion in space, or entropy density, due to dilution \(d\).

The correspondence of the enthalpy term \((\Delta H^{\text{c},0}/T)\) or entropy intensity term in Gibbs equation with a change of entropy density term labeled \(\Delta S_{\text{H}}\) can be explained. The entropy term \(\Delta S^{\text{c},0}\) can be calculated as

\[
\frac{-\Delta G^{\text{c},0}/T_{\text{max}}}{\Delta S_{\text{H}}}
\]

(25)
where \(T_{\text{max}}\) is the temperature at which \((\Delta H^{\text{c},0}/T)\) \(\to 0\), obtained by extrapolation to \((1/T) = 0\) of van’t Hoff equation. According to the second law and Carnot cycle, the term \((\Delta H^{\text{c},0}/T)\) represents thermal entropy transferred to the environment in an irreversible process. Within Gibbs equation, however, the thermal entropy term \((\Delta H^{\text{c},0}/T)\) is transformed and measured as an equivalent configuration entropy density term

\[
(\Delta H^{\text{c},0}/T) = \Delta S_{\text{H}}
\]

(26)
This entropy density term \(\Delta S_{\text{H}}\) is increasing when the temperature is decreasing, with the consequence that the equilibrium constant \(R \ln K\), which is an entropy density function, is increasing at low temperature. Hence the total entropy is increasing as well. Equation 26 represents a further example of ergodic property, based on an equivalence between entropy intensity (thermal) and entropy density (configuration). The equivalence

\[
-\Delta H^{\text{c},0} d(1/T) = R \ln d_{\text{A}}
\]

(27)
corresponds to equivalence of entropy (cf. Part II, Section 2\(^{11}\)).
The ergodic property of a system consists, experimentally, in the parallelism between the binding functions $R \ln K_{\text{dual}} = \{f(1/T)^*g(T)\}$ and $RT \ln K_{\text{dual}} = \{f(T)^*g(\ln T)\}$ and the binding quotients dependent on dilution $R \ln Q_K = \{f(1/d_{\text{id}(A)})^*g(T)\}$ and $RT \ln Q_K = \{f(d_{\text{id}(A)})^*g(\ln T)\}$, respectively (thermodynamic equivalent dilution (TED)) (cf. Part II, Section 3.1).  

### 2.3. Probability Space and Thermodynamic Space.

The equilibrium constant $K$ is related to free energy in thermodynamic space by

$$\Delta G^\circ / T = R \ln K$$  

(28)

that by transformation into an exponential becomes a monocentric partition function

$$K = \exp(-\Delta G^\circ / RT)$$  

(29)

in probability space. The exponential probability factor, is, therefore, represented by the equilibrium constant $K$, which has been shown, in analogy with $Q_K$, to be homologous with ideal calculated dilution $d_{\text{id}(A)}$. This means that state probability too is homologous with dilution.

Altogether, eqs 28 and 29 represent the connection between probability (or dilution) space and thermodynamic space. By analogy, we can move from Gibbs equation (eq 22), in thermodynamic space, to an exponential expression in probability space

$$\exp(-\Delta G^\circ / RT) = \exp(-\Delta H^\circ / RT) \cdot \exp(\Delta S^\circ / R)$$  

(30)

This equation tells us that the free-energy probability factor $\exp(-\Delta G^\circ / RT)$ can be factorized into a product of two probability factors, namely, $\exp(-\Delta H^\circ / RT)$, which depends on the reaction enthalpy of the thermodynamic space, and $\exp(\Delta S^\circ / R)$, which depends on the reaction entropy of the thermodynamic space.

We have shown that, following the ideas of Lumry,\(^1,2\) notwithstanding his pessimistic opinion on the experimental accessibility of thermal and motive components of the thermodynamic functions, we have calculated separated thermal and motive entropy as well as thermal and motive enthalpy, respectively. We have found (cf. Part II, Section 3) that the observed enthalpy $\Delta H_{\text{dual}}$ determined as derivative in $\partial(1/T)$ of the binding function $R \ln K_{\text{dual}} = \{f(1/T)^*g(T)\}$ can be represented in the thermodynamic space as

$$\Delta H_{\text{dual}} = \Delta H_0 + \Delta C_{p,\text{hydr}} T$$  

(31)

where $\Delta C_{p,\text{hydr}}$ hydrophobic heat capacity, is a constant independent of temperature $T$. On the other hand, we have found that the observed entropy $\Delta S_{\text{dual}}$ determined as a derivative in $\partial T$ of the binding function $RT \ln K_{\text{dual}} = \{f(T)^*g(\ln T)\}$ can be represented in the thermodynamic space by

$$\Delta S_{\text{dual}} = \Delta S_0 + \Delta C_{p,\text{hydr}} \ln T$$  

(32)

where the slope $\Delta C_{p,\text{hydr}}$ is numerically equal to the slope of the enthalpy function $\Delta H_{\text{dual}}$ for the same compound. We have obtained the expressions in eqs 31 and 32 by applying the extrapolation of $\Delta H_{\text{dual}}$ to $T = 0$ and of $\Delta S_{\text{dual}}$ to $\ln T = 0$, respectively, by taking advantage of constant $\Delta C_{p,\text{hydr}}$. We have identified the intercepts $\Delta H_0$ and $\Delta S_0$ as the motive components $\Delta H_{\text{mot}}$ and $\Delta S_{\text{mot}}$, respectively, foreseen but not calculated by Lumry. The constancy of heat capacity $\Delta C_{p,\text{hydr}}$ is necessary by both analytical geometry and chemistry constraints (cf. Part II, Section 3).\(^2\) We can write, therefore

$$\Delta H_{\text{dual}} = \Delta H_{\text{mot}} + \Delta H_{\text{th}}$$  

(33)

$$\Delta S_{\text{dual}} = \Delta S_{\text{mot}} + \Delta S_{\text{th}}$$  

(34)

The accuracy of the extrapolation procedure, based on $\Delta C_{p,\text{hydr}}$ constant, has been confirmed by the successive self-consistent results (cf. Part III),\(^1,2\) calculated for both motive and thermal components in every element of the many reactions examined.

Equation 34, whereby entropy $\Delta S_{\text{dual}}$ experimentally determined, is the result of a summation of entropy density $\Delta S_{\text{mot}} = \Delta S_{\text{dual}}$, with entropy intensity ($\Delta S_{\text{th}} = \Delta S_{\text{dual}}$), is a further proof of ergodic property of these systems. The ergodic hypothesis assumes the equivalence of configuration changes of entropy (entropy density), depending on space variables, with thermal changes of entropy, depending on time variables (entropy intensity) (i.e., the sum $\Delta S_{\text{dual}} = \Delta S_{\text{dual}} + \Delta S_{\text{dual}}$ is valid) (see Part II, Section 2). Each function in thermodynamic space yields an exponential function in probability space, as shown in Table 2. Therefore, from eqs 33–34 of the thermodynamic space, we can pass to the probability factors in probability (dilution) space

$$\exp(-\Delta H_{\text{dual}} / RT) = \{\exp(-\Delta H_{\text{mot}} / RT) \exp(-\Delta H_{\text{th}} / RT)\}$$  

(35)

and

$$\exp(\Delta S_{\text{dual}} / R) = \{\exp(\Delta S_{\text{mot}} / R) \exp(\Delta S_{\text{th}} / R)\}$$  

(36)

The probability factors of eqs 35–36 can be grouped together in a unique product partition function at dual structure

$$\{(\exp(-\Delta H_{\text{mot}} / RT))(\exp(\Delta S_{\text{mot}} / R))\} \cdot \exp(-\Delta H_{\text{th}} / RT)$$

$$\exp(\Delta S_{\text{th}} / R) = \{(\exp(-\Delta H_{\text{dual}} / RT))\exp(\Delta S_{\text{dual}} / R)\}$$

$$\exp(-\Delta G_{\text{dual}} / RT) = K_{\text{dual}}$$  

(37)

The dual-structure partition function $\{\text{DS-PF}\}$ of eq 37, valid for every hydrophobic hydration reaction, results to be the product of two distinct partition functions: a motive partition function $\{\text{M-PF}\} = \{f(T,d_{\text{id}(A)})\}$ multiplied by a thermal partition function $\{\text{T-PF}\} = g(T)$. $\{\text{M-PF}\}$ is of concern of the reacting solute and can give origin to configuration changes of entropy, i.e., changes of entropy density, whereas $\{\text{T-PF}\}$, concerning the solvent, can produce thermal changes of entropy, i.e., changes of entropy intensity.

The motive thermodynamic functions enthalpy ($\Delta H_{\text{mot}}$) and entropy ($\Delta S_{\text{mot}}$) are referred, in fact (see Part II, Section 4),\(^1\) to a reacting mole ensemble (REME), where the difference $\Delta H$ between levels is on the mole scale a multiple of $RT$ and $\Delta S$ is on the mole scale of the order of multiples of $R$ ($R = 8.314 J K^{-1} mol^{-1}$; capital M = mole). The mole ensemble is constituted by few elements (molecules). The variability of $S$ is, at constant temperature, of the order of $R$ times the differential of logarithm of reciprocal concentration, expressing the molar solvent-to-solute ratio (see eq 3).

The thermal functions enthalpy, $\Delta H_{\text{th}}$ and entropy, $\Delta S_{\text{th}}$, concerning the solvent, are referred to a nonreacting mole ensemble (NoremE; small $m$ = molecule), which is characterized by enthalpy levels very narrowly spaced, with interlevel separation of the order of magnitude $k_B T$ on the mole scale, where $k_B$ is the Boltzmann constant ($k_B = 1.3806 \times 10^{-23} J K^{-1}$). NoremE is composed of an extremely large number of elements (molecules). NoremE is independ-
Table 2. Relationships between Thermodynamic Space and Probability Space

\[
\begin{align*}
\text{thermodynamic space} & \quad | \quad \text{probability space} \\
-\Delta G^\circ / T & = K = \exp(-\Delta S_{\text{th}} / R) \\
-\Delta \text{G}_{\text{dual}} / T & = \Delta C_{\text{p,hydr}} dT = \Delta S_{\text{dual}} + \Delta S_{\text{hydr}} + \Delta S_{\text{th}} \\
\Delta C_{\text{p,hydr}} & = \exp((\Delta S_{\text{hydr}} / R)) \\
\Delta S_{\text{hydr}} & = \exp((\Delta S_{\text{hydr}} / R)) \\n\Delta S_{\text{th}} & = \exp((\Delta S_{\text{th}} / R)) \\
\text{Nonreacting molecule ensemble (NoremE)}: S & \equiv \Delta S_{\text{th}}
\end{align*}
\]

ent of concentration or dilution so that only thermal changes of entropy intensity can be produced (cf. \(dS_{\text{hydr}} = C_p d\ln T\) in eq 7).

We set a thermal probability factor referred to a NoremE

\[\exp(-\Delta H_{\text{th}} / RT) \exp(\Delta S_{\text{th}} / R) = 1\] (38)

The thermal functions in the thermodynamic space show special properties. By introducing the explicit values of the differentials, we obtain

\[dH_{\text{th}} = \Delta C_{\text{p,hydr}} dT\] (39)

and

\[dS_{\text{th}} = \Delta C_{\text{p,hydr}} d\ln T\] (40)

If we calculate the integral of eq 39 and then divide by \(T_{up}\), we obtain

\[
\left(1 / T_{up}\right) \int_0^{T_{up}} \Delta C_{\text{p,hydr}} dT = \Delta H_{\text{th}} / T_{up}
\]

\[
\approx \int_1^{T_{up}} \Delta C_{\text{p,hydr}} d\ln T = \Delta S_{\text{th}}
\]

where \(\Delta S_{\text{th}}\) is clearly identical to the result of the integration of eq 40. Therefore, we confirm that

\[\Delta H_{\text{th}} / T = \Delta S_{\text{th}}\] (42)

conform to eq 38 is a property of any thermal partition function \(\{T-PF\}\). We obtain the relation for thermal free energy

\[-\Delta G_{\text{th}} / T \equiv -\Delta H_{\text{th}} / T + \Delta S_{\text{th}} = 0\] (43)

which is invariably zero in accordance with eqs 38 and 42. This result is in contrast with the formula widely reported in the literature (cf. eq 79 in Part. II, Section 8 and citations therein), whereby the thermal free energy is erroneously stated to be different from zero.

Therefore, we can set the probability thermal factor as

\[\exp((\Delta C_{\text{p,hydr}} \ln T / R)) \exp((\Delta C_{\text{p,hydr}} \ln T / R) = 1\] (44)

where \((-\Delta H_{\text{th}} / T) \approx -\Delta C_{\text{p,hydr}} \ln T\) and \(\Delta S_{\text{th}} = \Delta C_{\text{p,hydr}} \ln T\)

In conclusion, the division of the thermodynamic functions into two parts, motive and thermal, proposed by Lumry (and numerically calculated by us for all of the many compounds examined), is a consequence of the special dual-structure partition function \(\{DS-PF\}\) (cf. eq 37), valid for every hydrophobic hydration process.

The introduction of the dual-structure partition function \(\{DS-PF\} = \{M-PF\} \cdot \{T-PF\}\) has been inspired by the suggestion of Lumry who proposed that the experimental functions \(\Delta H_{\text{dual}}\) and \(\Delta S_{\text{dual}}\) in hydrophobic hydration were separated into two parts, respectively, \(\Delta H_{\text{dual}} = \Delta H_{\text{mot}} + \Delta H_{\text{therm}}\) and \(\Delta S_{\text{dual}} = \Delta S_{\text{mot}} + \Delta S_{\text{therm}}\). Because a sum of exponent means a product of exponentials, we proposed the exponential product of eq 37. The thermal exponential is subject to the condition \(\exp((\Delta G_{\text{therm}} / T) = \zeta_{\text{th}} = 1\), with \((-\Delta G_{\text{therm}} / T) = 0\). This condition has been considered as indicative of a system at constant potential. The solvent, in a very diluted solution, is in excess and it does not change its concentration i.e., it is at constant potential. Therefore, the thermal function is suited to represent the properties of the solvent. The dual structure of the partition function corresponding to the biphasic composition of the system is
confirmed by the curved shape of the binding functions \( R \ln K_{\text{dual}} = \{f(1/T)g(T)\} \) and \( RT \ln K_{\text{dual}} = \{f(T)g(T)\} \) obtained by mathematical development of the dual-structure partition function, the binding functions result to be convoluted functions, whereby the primitive \( f(T) \) or \( f(T) \) of the motive component, which is linear, is modified by the secondary \( g(T) \) function, either \( g(T) \) or \( g(T) \) of the thermal component. We can identify the thermal partition function \( \{T-PF\} \) or \( g(T) \) as the partition function of the nonreacting solvent and the motive partition function \( \{M-PF\} = f(1/T,1/d_{\text{dil}}A) \) or \( f(T,1/d_{\text{dil}}A) \) as the partition function of the reacting solute.

**2.4. Ergodic Algorithmic Model (EAM).** The ergodic algorithmic model (EAM), obtained by the mathematical development of the dual-structure partition function \( \{DS-PF\} \) of eq 37, is based on a complex network of mathematical relationships connecting the whole set of experimental thermodynamic data (see Part II, Table 3).

**2.4.1. Motive and Thermal Ensembles.** We express the total statistical probability of the thermodynamic state of the system \(^{18}\) in analogy with eq 37 by

\[
\exp(-\Delta H_{\text{dual}}/RT) \exp(\Delta S_{\text{dual}}/R) = \exp(-\Delta G_{\text{dual}}/RT) = K_{\text{dual}}
\]

where \( K_{\text{dual}} \) is the observed equilibrium constant, \( \Delta H_{\text{dual}} \) is the observed reaction enthalpy, and \( \Delta S_{\text{dual}} \) is the observed entropy change. The formation constant \( K_{\text{dual}} \) has been demonstrated to be homologous with a partition function of statistical thermodynamics (cf. Table 1). At constant temperature, reciprocal concentration (i.e., dilution) in probability space is a measure of both formation constant and partition function.

The reciprocal concentration is related to the configuration changes of entropy density (cf. eq 3). The exponential factor \( \exp(-\Delta G_{\text{dual}}/RT) \) of the topological probability space, therefore, is homologous with the topological space of experimental reciprocal concentrations or dilutions.

An equilibrium constant is not the only probability parameter suitable to monitor the equilibrium conditions of the system: in the solubility of gases or liquids, the parameter is saturation concentration; in micelle formation, the parameter is critical micelle concentration; in protein denaturation, the parameter is the denaturation quotient \( Q_{\text{dual}} \) and so on.

In Table 3, we can note the correspondence between product probability functions (block C) and observed probability functions (block D), thus showing how the functions of the ergodic algorithmic model (EAM) conform to the observed thermodynamic functions.

As shown in eq 64, the thermal probability factor (or thermal partition function \( \{T-PF\} \)) is multiplied by a motive probability factor (or motive partition function, \( \{M-PF\} \)). The latter function is referred to a reacting mole ensemble (REME) (see Part II, Section 4) \(^{11}\) with equilibrium constant \( K_{\text{mot}} \)

\[
\exp(-\Delta H_{\text{mot}}/RT) \exp(\Delta S_{\text{mot}}/R) = \exp(-\Delta G_{\text{mot}}/RT) = K_{\text{mot}}
\]

Thus, we obtain a total dual-structure product probability factor \( \{DS-PF\} \)

\[
\{\exp(-\Delta H_{\text{mot}}/RT) \exp(\Delta S_{\text{mot}}/R)\}
\]

\[
= K_{\text{mot}} \cdot \exp(-\Delta H_{\text{dil}}/RT) \exp(\Delta S_{\text{dil}}/R)
\]

\[
= K_{\text{dual}}
\]

The total probability factor \( K_{\text{dual}} \) is the product of two partition functions: \( K_{\text{dual}} = K_{\text{mot}} \cdot \exp(-\Delta H_{\text{dil}}/RT) \exp(\Delta S_{\text{dil}}/R) \)

\[
= K_{\text{dual}}
\]

| Table 3. Ergodic Algorithmic Model (EAM) from Probability Space to Thermodynamic Space |
|---------------------------------|-------------------|-------------------|
| A                              | motive function    | units             |
| probability space              | \exp(-\Delta H_{\text{dual}}/RT) \exp(\Delta S_{\text{dual}}/R) = \exp(-\Delta G_{\text{dual}}/RT) = K_{\text{dual}} | eq 45           |
| thermodyn. space               | \begin{align*}
R \ln K_{\text{dual}} &= \{f(1/T)g(T)\} \exp(-\Delta H_{\text{dual}}/RT) + \{g(T)\} \exp(\Delta S_{\text{dual}}/R) = \exp(-\Delta G_{\text{dual}}/RT) \\
RT \ln K_{\text{dual}} &= \{f(T)g(T)\} \exp(-\Delta H_{\text{dual}}/RT) + \{g(T)\} \exp(\Delta S_{\text{dual}}/R) = \exp(-\Delta G_{\text{dual}}/RT)
\end{align*} | eq 46   |
| B                              | thermal function   | units             |
| probability space              | \exp(-\Delta H_{\text{dual}}/RT) \exp(\Delta S_{\text{dual}}/R) = \exp(-\Delta G_{\text{dual}}/RT) = K_{\text{dual}} | eq 47   |
| thermodyn. space               | \begin{align*}
(\Delta H_{\text{dil}}) &= \Delta C_{\text{ph}\text{dil}} T \\
(\Delta H_{\text{dil}}) &= \Delta C_{\text{ph}\text{dil}} T \\
\Delta S_{\text{dil}} &= \Delta C_{\text{ph}\text{dil}} T \\
(\Delta H_{\text{dil}}) + \Delta S_{\text{dil}} &= 0
\end{align*} | eq 48   |
| C                              | product function   | units             |
| probability space              | \exp(-\Delta H_{\text{dual}}/RT) \exp(\Delta S_{\text{dual}}/R) = \exp(-\Delta G_{\text{dual}}/RT) = K_{\text{dual}} | eq 49   |
| thermodyn. space               | \begin{align*}
R \ln K_{\text{dual}} &= \{f(1/T)g(T)\} \exp(-\Delta H_{\text{dual}}/RT) + \{g(T)\} \exp(\Delta S_{\text{dual}}/R) = \exp(-\Delta G_{\text{dual}}/RT) \\
RT \ln K_{\text{dual}} &= \{f(T)g(T)\} \exp(-\Delta H_{\text{dual}}/RT) + \{g(T)\} \exp(\Delta S_{\text{dual}}/R) = \exp(-\Delta G_{\text{dual}}/RT)
\end{align*} | eq 50   |
| D                              | observed function  | units             |
| probability space              | \exp(-\Delta H_{\text{dual}}/RT) \exp(\Delta S_{\text{dual}}/R) = \exp(-\Delta G_{\text{dual}}/RT) = K_{\text{dual}} | eq 51   |
| thermodyn. space               | \begin{align*}
R \ln K_{\text{dual}} &= \{f(1/T)g(T)\} \exp(-\Delta H_{\text{dual}}/RT) + \{g(T)\} \exp(\Delta S_{\text{dual}}/R) = \exp(-\Delta G_{\text{dual}}/RT) \\
RT \ln K_{\text{dual}} &= \{f(T)g(T)\} \exp(-\Delta H_{\text{dual}}/RT) + \{g(T)\} \exp(\Delta S_{\text{dual}}/R) = \exp(-\Delta G_{\text{dual}}/RT)
\end{align*} | eq 52   |

**References**

1. ACS Omega, 2018, 3, 15043–15065.

**DOI:** 10.1021/acsomega.0b01685
0, whereas \( \zeta \) is the thermal partition function \((\text{T-PF})\) of the solvent, referred to a nonreacting molecule ensemble (Norem-E), with \((-\Delta G_{th}/RT) = 0\).

The curved binding functions (convoluted functions) obtained by developing \((\text{DS-PF})\) (see block C in Table 3) can be compared to the curved binding functions obtained by interpolation of the experimental data reported in a van’t Hoff plot (see block D in Table 3). The tangents \( \Delta H_{\text{dual}} \) and \( \Delta S_{\text{dual}} \) of the experimental binding functions interpolating the experimental data are calculated as a sum of two terms (ergodic) as shown in eqs 33 and 34, respectively. From the experimental data, we obtain the following expressions for enthalpy and entropy probability factors, respectively:

\[
\exp(-\Delta H_{\text{dual}}/RT) = \exp(-\Delta H_{\text{mot}}/RT) \exp(-\Delta H_{\text{th}}/RT)
\]

\[
\exp(\Delta S_{\text{dual}}/R) = \exp(\Delta S_{\text{mot}}/R) \exp(\Delta S_{\text{th}}/R)
\]

which clearly conform to eq 65.

### 2.4.2. Binding Functions

By passing to the logarithms of the partition functions, we move from probability space, homologous with the reciprocal concentration (dilution) space, to thermodynamic space, where we measure energy, enthalpy, and entropy \((\text{see Appendix B})\). We thus obtain the expressions presented in block C of Table 3. We note that, in Table 3, the observed binding functions

\[
R \ln K_{\text{dual}} = \{\Delta H_{\text{mot}} - \Delta H_{\text{th}}\} + \{\Delta S_{\text{mot}} + \Delta S_{\text{th}}\}
\]

\[
= -\Delta G_{\text{dual}}/T
\]

and

\[
RT \ln K_{\text{dual}} = \{-\Delta H_{\text{mot}} - \Delta H_{\text{th}}\} + T\{\Delta S_{\text{mot}} + \Delta S_{\text{th}}\}
\]

\[
= -\Delta G_{\text{dual}}
\]

are expressed in J K\(^{-1}\) mol\(^{-1}\) (entropy scale) and J mol\(^{-1}\) (enthalpy scale), respectively, thus confirming that they are in the thermodynamic space.

The two functions in eq 68 and in eq 69, respectively (see Appendix C), present diagrams with curvature, depending on the value of \(\Delta C_{\text{phym}}\). The thermal factor can be either of class A when \(\Delta C_{\text{phym}} > 0\) or of class B when \(\Delta C_{\text{phym}} < 0\). If \(\Delta C_{\text{phym}} > 0\), the binding functions (cf. Section 2.5.3) are monotonic convex at constant curvature amplitude, whereas if \(\Delta C_{\text{phym}} < 0\), the binding functions are monotonic concave at constant curvature amplitude. The curvature amplitude is a constant typical of each parabola and is inversely proportional to the constant heat capacity \(\Delta C_{\text{phym}}\). The thermal factor can be either of class A or from solute (WII) to solvent WI in all of the compounds of class A or from solute (WII) to solvent WI in all of the compounds of class B, respectively (see the definitions of the properties of class A and class B), \(\Delta C_{\text{phym}}\) results to be constant for chemical conditions also. Any phase transition from WI (solute) to WII (iceberg) is characterized, in fact, by constant entropy change equal to an enthalpy change divided by temperature, which means that heat capacity could be labeled as entropic function \(\Delta C_{\text{phym}} = \Delta H_{\text{phym}}/T = \Delta S_{\text{hym}}\).

The passage of state of pure water to form an iceberg is analogous to evaporation, although with its own characteristics, with \(\Delta S_{\text{fw}} = \Delta C_{\text{fw}} = 75.36\) J K\(^{-1}\) mol\(^{-1}\). The interpretation of \(\Delta S_{\text{fw}} = \Delta C_{\text{fw}}\) as a constant value of entropy change for a phase transition has the important implication that the existence of the phase transition WI (solute) \(\rightarrow\) WII (iceberg) (with \(n_{\text{w}} = 1\)) explains the abnormal high value of heat capacity of liquid water. This high value is inconsistent with a simple redistribution of energies over degrees of freedom of a nonreacting water molecule, as for usual interpretation of heat capacity. The phase transition of water, in fact, should take place even when we determine heat capacity of pure liquid water by calorimetry, thus giving a prominent contribution to heat capacity.
By passing to the logarithms as in eqs 68 and 69 and then differentiating, we obtain the relations collected in Table 4. In Table 4, we note that, by assuming \( \Delta C_{\text{ph}} \) constant and independent of temperature, we arrive at identical \( \Delta C_{\text{ph}} \) by both enthalpy route and entropy route. The same equality of \( \Delta C_{\text{ph}} \) from either enthalpy or entropy is obtained by treating the experimental data. We can conclude, therefore, that the experimental data conform to the ergodic algorithmic model (EAM) and contain in themselves \( \Delta C_{\text{ph}} \) constant, independent of temperature. We can thus confirm once again that the heat capacity \( \Delta C_{\text{ph}} \) is constant for both chemistry and analytical geometry constraints.

### 2.4.3. Characteristic Properties of Binding Functions

The relationships between experimental data, free energy, enthalpy, and entropy as expressed by the two binding functions \( R \ln K_{\text{dual}} = \{f(1/T)g(T)\} = -\Delta G_{\text{dual}}/T \) and \( RT \ln K_{\text{dual}} = \{f(T)g(1/T)\} = -\Delta G_{\text{dual}} \) are reported in Table 5 of Part II. Being \( \Delta C_{\text{ph}} \neq 0 \), the functions \( R \ln K_{\text{dual}} = \{f(1000/T)g(T)\} \) and \( RT \ln K_{\text{dual}} = \{f(T)g(1000/T)\} \) are curvilinear (Figure 2a,b) represented by second-degree polynomials (see Appendix C). It is worth mentioning that the convex curves in Figure 2a,b are referred to the experimental solubility data of helium in water. These kinds of processes concerning gases belong to class A of the hydrophobic hydration processes. If we calculate the derivatives \( \partial(R \ln K_{\text{dual}})/\partial(1/T) \) and \( \partial(RT \ln K_{\text{dual}})/\partial T \) and plot them against \( T \) and \( \ln T \), respectively, we obtain linear plots (Figure 3a,b). We remind that the indicated derivatives correspond to the tangents at any point of the curves in Figure 2a,b. The value of the tangent at the point indicated in the diagram \( R \ln K_{\text{dual}} = f(1/T) \) (cf. Figure 3a) corresponds to the value of \( -\Delta H_{\text{dual}} \) whereas the value of the tangent at any point in the diagram \( RT \ln K_{\text{dual}} = f(T) \) (cf. Figure 3b) corresponds to the value of \( \Delta S_{\text{dual}} \). In these diagrams, \( T_H \) is the temperature at which \( \Delta H_{\text{dual}} = 0 \) and \( \Delta S_{\text{dual}} \) at \( T_S \). According to the ergodic algorithmic model presenting curved binding functions, the apparent dual enthalpy (i.e., the experimental enthalpy) represented by the tangent of one binding function is composed of two terms, motive (i.e., entropy density) and thermal (i.e., entropy intensity) (cf. eq 33).

The motive enthalpy \( \Delta H_{\text{mot}} \) is independent of the temperature, whereas the thermal enthalpy \( \Delta H_{\text{th}} \) depends exclusively on the temperature with proportionality factor \( \Delta C_{\text{ph}} \).

\[
\Delta H_{\text{th}} = T \Delta C_{\text{ph}}
\]

(79)

At \( T_{10} \), the enthalpy is zero because

\[
\Delta H_{\text{mot}} + \Delta H_{\text{th}} = \Delta H_{\text{dual}} = 0
\]

(80)

therefore, by introducing eq 79 into eq 80, we can calculate

\[
\Delta H_{\text{mot}} = T_{10} \Delta C_{\text{ph}}
\]

(81)

In this equation, the extrapolation to \( T = 0 \) has been applied, and the extrapolation to \( \ln T = 0 \) will be applied in eq 84: these procedures are legal because \( \Delta C_{\text{ph}} \) is constant.

On the other hand, with the same arguments already used for enthalpy and \( T_{10} \), we can calculate the motive entropy

\[
\Delta S_{\text{mot}} = \Delta C_{\text{ph}} \ln T_S
\]

(82)

The relationships presented so far are referred to solubility data of argon that is a reaction belonging to class A with \( \Delta C_{\text{ph}} > 0 \) and iceberg formation. The condition \( \Delta C_{\text{ph}} > 0 \) characterizes the binding functions as convex.

Analogous relationships are valid for reactions belonging to class B, with iceberg reduction (see Appendix C). The curves in the diagrams of class B present a maximum instead of a minimum, i.e., are concave. This is shown in an application of these relationships referred to processes of class B (Figure 4a,b). Talhout et al.\(^{15} \) studied the binding affinity of a series of hydrophobically modified benzamidinium chloride inhibitors binding to trypsin, using isothermal calorimetry and molecular dynamic simulation techniques. The binding functions \( R \ln K_{\text{dual}} \) and \( RT \ln K_{\text{dual}} \) reported as the function of \( 1/T \) and \( T \), respectively, show curved plots, as expected for hydrophobic...
hydration processes (cf. Figure 4a,b) of class B. The affinity between ligand benzamidinium chloride and enzyme is due presumably to the formation of hydrophobic bonds as revealed by the negative values of the reaction heat capacity \( \Delta C_{p,\text{hydr}} \) (\( \Delta C_{p,\text{hydr}} < 0 \)). The condition \( \Delta C_{p,\text{hydr}} < 0 \) characterizes the functions as concave. The diagram \( (- \Delta G/T) = f(1/T) \) with a maximum confirms that we are dealing with a hydrophobic hydration process of class B. The tangent of the curve is

\[
\partial(-\Delta G_{\text{dual}}/T)/\partial(1/T) = -\Delta H_{\text{dual}} \tag{83}
\]

This tangent is variable with temperature

\[
\partial(\Delta H_{\text{dual}})/\partial T = \Delta C_{p,\text{hydr}} \tag{84}
\]

At the maximum, the tangent \( \Delta H_{\text{dual}} \) is nil

\[
\partial(-\Delta G_{\text{dual}}/T)/\partial(1/T) = 0 \tag{85}
\]

We define \( T_{50} \) as the temperature at which \( \Delta H_{\text{dual}} = 0 \).

Analogously, the plot \( (- \Delta G_{\text{dual}}) = f(T) \) is a curve with a maximum at a different temperature. The tangent to this curve is

\[
\partial(-\Delta G_{\text{dual}})/\partial T = \Delta S_{\text{dual}} \tag{86}
\]

In this case also, the tangent varies with temperature

\[
\partial(\Delta S_{\text{dual}})/\partial \ln T = \Delta C_{p,\text{hydr}} \tag{87}
\]

At the maximum, \( \Delta S_{\text{dual}} \) is nil

\[
\Delta S_{\text{dual}} = \partial(-\Delta G_{\text{dual}})/\partial T = 0 \tag{88}
\]

We define \( T_{50} \) as the temperature at which \( \Delta S_{\text{dual}} = 0 \). Talhout et al.\textsuperscript{13} report the data \( \Delta C_{p,\text{hydr}}, T_{50} \) and \( T_{100} \) concerning the binding curves of eight compounds (Table 5). The thermodynamic data of every compound in the list conform to the ergodic algorithmic model (EAM).

**2.4.4. Motive Functions Disaggregated as the Functions of**

**\( T_{50} \)**

The separation of the thermodynamic functions \( \Delta H_{\text{dual}} \) and \( \Delta S_{\text{dual}} \) into thermal and motive components as proposed by Lumry\textsuperscript{1,2} and calculated by us by applying the ergodic algorithmic model (EAM)

\[
\Delta H_{\text{dual}} = \Delta H_{\text{mot}} + \Delta H_{\text{th}}(T) \tag{89}
\]

and hence (Figure 5) an exact compensation between thermal and motive enthalpy

\[
0 = \Delta H_{\text{mot}} + \Delta H_{\text{th}}(T) \tag{91}
\]

**Figure 5. Calculation of \( \Delta H_{\text{mot}} \) from the data reported by Talhout et al.\textsuperscript{13} for the ligand benzamidinium chloride (class B).**

\[
\Delta H_{\text{mot}} = -\Delta C_{p,\text{hydr}} T_{100} \tag{92}
\]

By knowing \( \Delta C_{p,\text{hydr}} \) from TED, we can calculate \( \Delta H_{\text{mot}} \) (Figure 6). Analogously, for entropy in class B, we have

\[
\Delta S_{\text{dual}} = \Delta S_{\text{mot}} + \Delta S_{\text{th}}(T) \tag{93}
\]

**Figure 6. Calculation of \( \Delta S_{\text{mot}} \) from the data reported by Talhout et al.\textsuperscript{13} for the ligand benzamidinium chloride (class B).**

From \( \Delta C_{p,\text{hydr}} \) we can calculate for each compound, by applying TED (cf. eq 104 below)

\[
n_{w} = \Delta C_{p,\text{hydr}}/C_{p,w} \tag{94}
\]

where \( C_{p,w} = 75.36 \text{ J K}^{-1} \text{ mol}^{-1} \) is the heat capacity of liquid water.

The motive functions \( \Delta H_{\text{mot}} \) and \( \Delta S_{\text{mot}} \) calculated for a homogeneous set of compounds can be disaggregated by

| subst. R | \( \Delta C_{p,\text{hydr}} \) (J K \(^{-1} \text{ mol}^{-1} \)) | \( T_{50} \) (K) | \( \ln T_{50} \) | \( T_{100} \) (K) | \( \Delta S_{\text{mot}} \) (J K \(^{-1} \text{ mol}^{-1} \)) | \( \Delta H_{\text{mot}} \) (kJ mol \(^{-1} \)) | \( n_{w} \) |
|----------|-----------------|----------|-------------|----------|-----------------|-----------------|----------|
| H        | -400            | 317.15   | 5.759375    | 250.15   | 2303.75         | 100.06          | -5.31     |
| Me       | -420            | 319.15   | 5.765661    | 254.15   | 2421.578        | 106.74          | -5.57     |
| Et       | -603            | 315.15   | 5.753049    | 277.95   | 3469.088        | 167.60          | -8.00     |
| n-Pr     | -598            | 320.15   | 5.76879     | 277.05   | 3449.736        | 165.68          | -7.94     |
| i-Pr     | -419            | 336.15   | 5.817557    | 285.15   | 2437.557        | 117.97          | -5.56     |
| n-Bu     | -728            | 321.15   | 5.771908    | 285.15   | 4021.949        | 207.59          | -9.66     |
| n-Pent   | -632            | 328.15   | 5.793471    | 282.75   | 3661.474        | 178.70          | -8.39     |
| n-Hex    | -849            | 321.15   | 5.771908    | 285.15   | 4900.33         | 242.09          | -11.27    |
plotting them against $\xi = \ln l$ (Figure 7) of each compound of the set. The enthalpy interpolating function (Figure 7A) for

the set of ligands (substituted benzanilidinium chlorides) studied by Talhout et al.$^{13}$ is

$$\Delta H_{\text{mot}} = -19.61 + 23.406 \xi_w$$

(95)

that is coincident with the mean value of class B $\langle \Delta h_{\text{mol}} \rangle_b = +23.7 \pm 0.6$ kJ mol$^{-1}$ $\xi_w^{-1}$ (see Table 4a in Part III).$^{12}$

The entropy interpolating function (Figure 7B) in the same group of compounds is

$$\Delta S_{\text{mot}} = +5.8246 + 434.41 \xi_w$$

(96)

The slope is compared to the mean value of unitary entropy of class B $\langle \Delta h_{\text{mol}} \rangle_b = +432 \pm 4$ J K$^{-1}$ mol$^{-1}$ $\xi_w^{-1}$. The comparison of the unitary values calculated from the data of Talhout et al.$^{13}$ with the unitary values reported in Table 5 of Part III$^{12}$ demonstrates that the foundations of the model are sound. A constant value of $\Delta C_{\text{p,hydro}}$ independent of temperature, is a necessary property to construct the linear functions $\Delta H_{\text{dual}} = f(T)$ and $\Delta S_{\text{dual}} = f(\ln T)$ used by Talhout et al. Specifically, the numbers $\xi_w = \ln l$ calculated from the slopes of the linear thermal functions can be successfully employed to disaggregate the corresponding motive functions in a homologous series of compounds. All of the numerical values of the thermodynamic functions are self-consistent. The separation of the dual functions into thermal and motive components leads to coherent numerical results. These proofs represent further validations of the EAM model itself because the statistical analysis is indisputable (cf. Section 5 in Part III).$^{12}$

2.4.5. Ergodic Properties. We can complete the description of the ergodic algorithmic model (EAM) by introducing the formal mathematical expression of ergodicity of hydrophobic hydration systems.

The analysis of the binding functions of the ergodic algorithmic model (EAM) assumes that the function entropy is a measure of “energy dispersal” in the system. The concept of “dispersion” can be clearly grasped by analogy with concentration $x_A$ of species A and its reciprocal ideal dilution $d_{id,A} = 1/x_A$. $d_{id,A}$ is a measure of the volume in which 1 mol is dispersed (see Appendix C). If we suppose to measure a quantity $\varepsilon$ of energy density, the dispersion of energy is given by $1/\varepsilon$. The thermodynamic function entropy $S$ is proportional to such energy dispersal, $1/\varepsilon$. The dispersal of energy can take place by two different mechanisms: (a) energy dispersion in time and (b) energy dispersion in space. The dispersion in time depends on the velocity of the molecules through the variable temperature $T$, whereas the dispersion in space depends on variable concentration $x_A$ or better on variable dilution $d_{id,A} = 1/x_A$. The dispersion of energy in time increases with temperature $T$ because the temperature is proportional to the squared mean velocity of the particles. In fact, if the molecule is running faster, then the energy carried by the molecule flows more rapidly through the cell, increasing the dispersion of energy over a longer path in time unit. The dispersion of energy in time is measured by the squared mean sojourn time, $\tau_w^{-1}$. The sojourn time $\tau_i$ of $i$th particle is the time spent by one molecule to run the length unit ($\tau_i = 1/l_i$). As for dispersion in space, if we imagine that each molecule is carrying an amount of energy, when we dilute the molecules, we dilute, at the same time, the energy associated to each molecule, thus increasing the dispersion of energy in space. We call entropy intensity the dispersion of energy in time as the function of temperature $T$, whereas we call entropy density the dispersion of energy in space as the function of dilution $d_{id,A}$. We obtain in such a way for entropy density

$$\langle dS_{\text{dual}} \rangle_T = (-R \ln x_A)_T$$

(97)

and for entropy intensity

$$\langle dS_{\text{dual}} \rangle_{xA} = (C_P \ln T)_{xA}$$

(98)

The ergodic hypothesis assumes that the variability in time of entropy or entropy intensity in a thermodynamic system is equivalent to the variability in space of entropy or entropy density: entropy intensity and entropy density can be summed up (ergodicity) because both are parameters of energy dispersal. We have experimentally verified that in every hydrophobic hydration system, the equivalence can be experimentally found as thermal equivalent dilution (TED): variability of $R \ln K$ as the function of $1/T$ is parallel to the variability of $R \ln K$ as the function of $1/d_{id,A}$.

The activity of a species A is set as

$$a_A = x_A \Phi$$

(99)

where $x_A$ is the concentration of A in molar fraction and $\Phi = T^{-c_w}/R$ is the thermal factor. The thermal equivalent dilution (TED) principle becomes

$$n_w(R \ln d_{id,A})_T = n_w(C_p \ln T)_{xA}$$

(100)

whereby a change of entropy density, as obtained from the motive partition function $\{M-PF\} = \{f(1/T)^*f(d_{id,A})\}$ is experimentally determined by measuring a parallel change of entropy intensity by means of the binding function $R \ln K_{dual} = \{f(1/T)^*g(T)\}$ at constant $d_{id,A}$. The potential distribution theorem (PDT),$^7$ based on a monocentric partition function, without distinction of entropy density and entropy intensity, is inconsistent with the dual composition of biphasic hydrophobic hydration systems (Table 6).

On the other hand, the development of the theorem PDT called “quasi-chemical approximation” (improperly defined approximation) is valid because it considers the different distribution types of the two phases: molecule statistical
distribution of the solvent (entropy intensity) and mole bimodal distribution of the reacting solute (entropy density) (cf. Part II, Section 10.b).¹¹

2.5. Hydrophobic Heat Capacity, ΔCₚ,hydr.  
2.5.1. ΔCₚ,hydr. Equilibrium Constant, and TED. The ergodic algorithmic model (EAM) is completed by the relationships between binding functions and activities of the reacting species. The TED method based on the assumption of the validity of the so-called “ergodic hypothesis” (see Part II, Section 2)¹¹ has appeared as a potent experimental tool to evaluate the number nₓ of moles of water Wₓ involved in each reaction. The number nₓ can be either positive or negative. The number nₓ will be positive in the processes of class A and negative in the processes of class B. The number nₓ as determined by TED, was assumed at first as being simply proportional to the volume of the incoming solute and dependent on a generic concentration of water molecules W. During subsequent researches, however, it has been proved to correspond to the real number nₓ = lnₓ l of water clusters Wₓ involved in each reaction (cf. eqs 98 and 99) and as such we can consider it. This has opened the way to determine the real number x w whereby the water units Wₓ enter as reactant in every hydrophobic hydration process. The absolute value w = lnₓ l was adopted because in a group of reactions (class B) nₓ is negative and the introduction of the absolute value transfers any change of sign to the associated thermodynamic quantity, with meaningful thermodynamic and molecular implications.

The relationship between hydrophobic heat capacity Cₚ,hydr and number x w = lnₓ l through TED can be found by recalling that eq 95 can be written as a double derivative in ln T

\[ -\frac{\partial^2 (-\Delta G^0 / T)}{\partial (1 / T)} = \frac{\partial^2 (-\Delta G^0 / T)}{\partial (\ln T)^2} = \Delta C_{P,hydr} \]  

(101)

We can set a dissociation constant, valid for class A

\[ K_{diss} = (a_x)(a_{WII})^{x_w}/(a_y)^{1-x_w} \]  

(102)

where aₓ, aₓ WII, and aₓ indicate activities of the species. The changing of the equilibrium constant at different temperatures can be represented by a series

\[ R \ln K = R \ln K_0 + \frac{\partial (R \ln K)}{\partial (\ln T)} \ln T + \frac{\partial^2 (R \ln K)}{\partial (\ln T)^2} (\ln T)^2 \]  

(103)

where R ln K₀ = (−ΔGₓ μ₀/T). The second moment of the distribution is represented in eq 99. Alternatively, the second moment can be written as the first derivative of the differential changes δ₁ ln aₓ, δ₁ ln aₓ, δ₁ ln aₓ WII induced by the first moment (derivative)

\[ \frac{\partial}{\partial (\ln T)} \left[ \frac{\partial (R \ln K)}{\partial (\ln T)} \right] = \frac{\partial (R \delta_1 \ln a_x)}{\partial (\ln T)} - \frac{\partial (R \delta_1 \ln a_y)}{\partial (\ln T)} + \frac{\partial (R \delta_1 \ln (a_{WII})^{x_w})}{\partial (\ln T)} = \Delta C_{P,hydr} \]  

(104)

Because changes δ₁ ln aₓ and δ₁ ln aᵧ compensate each other, their contribution is null

\[ \partial R \delta_1 \ln a_x/\partial (\ln T) - \partial R \delta_1 \ln a_y/\partial (\ln T) = 0 \]  

(105)

and then the hydrophobic heat capacity is expressed by

\[ R \ln (a_{WII})^{x_w}/(\ln T) = \Delta C_{P,hydr} \]  

(106)

The TED principle for a species X with activity aₓ states

\[ -n_R \ln a_x = n C_{p,X} d(\ln T) = dS_x \]  

(107)

where Cₓ pX is the heat capacity and Sₓ is the configuration change of entropy of X.

Therefore, we obtain for the factor (a_{WII})^{x_w} of water Wₓ the following equality

\[ \delta (\xi_w, R \ln a_{WII})/\partial (\ln T) = \xi_w C_{p,w} = \Delta C_{P,hydr} \]  

(108)

where Cₓ pW = 75.36 J K⁻¹ mol⁻¹ is the isobaric heat capacity of liquid water.

Alternatively, by changing sign to x w we set an association constant, valid for class B

\[ K_{assoc} = (a_y)^{1-x_w}(a_{WII})^{-x_w} \]  

(109)

and obtain

\[ -\xi_w C_{p,w} = \Delta C_{P,hydr} \]  

(110)

which represents the curvature in the binding functions of class B that present a maximum. There is a relationship between change in virtual dilution and curvature analogous to that between ΔCₓ p,hydr and curvature. This means that the curvatures depend on the number w = lnₓ l. The determination of the pseudostoichiometric number nₓ w can be achieved by the variation of virtual dilution δ(−nₓ ln Wₓ/∂(ln T ≠ 0) and hence the variation of the equilibrium constant brought about by the Lambent thermal energy factor Φ of water molecules Wₓ when the temperature is changing. The reaction coefficient x w is qualified as “pseudo” because it is in general noninteger, indicating the ratio between volume of incoming moity and volume of cluster Wₓ. We can verify that the variation of the virtual dilution dₓ Wₓ of water Wₓ (d_{WII} = 1/α_{WII}) is the unique cause of the curvature (as convoluted function, cf. Figure 9) in the plot (−ΔGₓ μ₀/T = f(1/T), as it is ΔCₓ p,hydr.

The coherence and self-consistency of the numerical results obtained by applying TED can be considered as the experimental proofs of the validity of the “ergodic theory or ergodic hypothesis”: we can now speak of “ergodic properties” of thermodynamic systems, dismissing the word “hypothesis”. The energy fluxes in intensity (thermal) and density (configurational) entropy are equal (cf. Figure 5 in Part II).¹¹

2.5.2. ΔCₓ p,hydr and Phase Transition in Water Wₓ. The hydrophobic hydration processes are characterized by large values of heat capacity ΔCₓ p,hydr. The values of heat capacity

\[ \Delta C_{P,hydr} = \Delta H_{dual}/T \]  

(111)
are calculated in general by plotting the values of the observed enthalpy change $\Delta H_{\text{dual}}$ against $T$ and then by interpolating the points by a straight line of equation

$$\Delta H_{\text{dual}} = \Delta H_0 + T \cdot \Delta C_{p,\text{hydr}}$$  \hfill (112)

where $\Delta C_{p,\text{hydr}}$ is the hydrophobic heat capacity. The same rule holds for both calorimetric and van’t Hoff enthalpy. There has been, however, some debate whether this equation is exactly valid or only approximately valid. The question, in other words, is whether the heat capacity is constant for analytical geometry constraint. The question, in other words, is whether the heat capacity is constant for analytical geometry constraint. The equality of the coefficients in the two diagrams is possible only if $\Delta C_{p,\text{hydr}}$ is constant and independent of temperature, in accordance with the equal curvature amplitudes of both binding functions.

All of these findings representing the experimental evidence that $\Delta C_{p,\text{hydr}}$ is independent of the temperature actually conform to the ergodic algorithmic model (EAM).

Another significant characteristic of $\Delta C_{p,\text{hydr}}$ is that in certain processes, we find $\Delta C_{p,\text{hydr}} > 0$ and in others, $\Delta C_{p,\text{hydr}} < 0$. Even in the compounds with negative $\Delta C_{p,\text{hydr}}$, we find the identity of the coefficients $\Delta C_{p,\text{hydr}}$ in the diagrams $\Delta H_{\text{dual}} = \Delta H_0 + g(T)$ and $\Delta S_{\text{dual}} = \Delta S_0 + g(T)$. We have defined the processes with positive heat capacity ($\Delta C_{p,\text{hydr}} > 0$) as belonging to class A and those with negative slope ($\Delta C_{p,\text{hydr}} < 0$) as belonging to class B. The hydrophobic heat capacity $\Delta C_{p,\text{hydr}} = \pm \xi_w C_{p,w}$ (where $C_{p,w} = 75.36$ J K$^{-1}$ mol$^{-1}$ is the isobaric heat capacity of liquid water) depends on the stoichiometry of the reaction with phase transition between water components $W_I, W_{II}$. In class A, the reaction with phase transition is

$$A\{\xi_w W_I(\text{solvent}) \rightarrow \xi_w W_{II}(\text{iceberg})\}$$

whereas in class B, the reaction with opposite phase transition is

$$B\{ - \xi_w W_{II}(\text{iceberg}) \rightarrow \xi_w W_I(\text{solvent})\}$$

with iceberg reduction. It is worth noting that iceberg formation in class A and iceberg reduction in class B, respectively, produce changes of the thermodynamic properties of the solute. As any entropy change, in analogy with Trouton’s law, $\Delta C_{p,\text{hydr}}$ could be labeled as $\Delta H_{p,\text{hydr}}/T$. For any phase-transition entropy, $\Delta C_{p,\text{hydr}} = \Delta H_{p,\text{hydr}}/T$ is constant, independent of the temperature. In general, the phase transition takes place for each compound at a fixed temperature; in these cases, however, the condition holds at all temperatures of measurement.

The curvature amplitude of the binding functions is inversely proportional to the constant hydrophobic heat
capacity $\Delta C_p$ as confirmed by an analysis (see Section 2.5.3) of the experimental data concerning the denaturation of proteins.

2.5.3. $\Delta C_p$ and Curvature Amplitude of Binding Functions. Coming specifically to protein denaturation, belonging to class A, we have shown that in the plot $(-\Delta G_{dual}/T) = f(1/T) g(T)$, the curves obtained at different pH and different temperatures present the same curvature. In fact, the tangent of the function is

$$\frac{\partial (-\Delta G_{dual}/T)}{\partial (1/T)} = -\Delta H_{dual}$$

(114)

The family of tangents of a curve (i.e., values of $\Delta H_{dual}$) calculated at different values of the abscissa are straight lines of variable slopes. By calculating the derivative of $\Delta H_{dual}$ with respect to the variable $T$, we obtain the heat capacity

$$\frac{\partial (\Delta H_{dual})}{\partial T} = \Delta C_p$$

(115)

We note that if the original function $(-\Delta G_{dual}/T) = f(1/T)$ were a straight line, as in normal van’t Hoff plots, the derivative of eq 110 would be a constant at any temperature and the derivative in eq 111 would be zero. Therefore, if $\Delta H_{dual}$ were constant at different temperatures, we should have $\Delta C_p = 0$. On the other hand, if the function $(-\Delta G_{dual}/T) = f(1/T)$ is a curve, then $\frac{\partial (\Delta H_{dual})}{\partial T} \neq 0$, and hence we can conclude that the function is not a simple function $f(1/T)$, rather it will come out to be a convoluted function $(-\Delta G_{dual}/T) = f(1/T) g(T)$. If by deriving further eq 84, we obtain $\frac{\partial (\Delta C_p)}{\partial T} = 0$, then we conclude that $\Delta C_p$ is constant. Therefore, the curvature amplitude of van’t Hoff plots is constant as well. The reciprocal value of $\Delta C_p$ is a measure of the curvature amplitude of the function $(-\Delta G_{dual}/T) = f(1/T) g(T)$. The experimental data for DMS derivative of chymotrypsinogen are reported in Figure 8A. If the curvature of van’t Hoff plot is constant and independent of $T$, a vertical displacement downward, simply by changing pH, of the experimental values of log $Q_{\text{den}}$ gets curves. Labeled a–d in Figure 8A, without altering the curvature. From a single curve, therefore, one can obtain sections displaced downward to bring every section to values of log $Q_{\text{den}}$ around zero. The concentration quotient $Q_{\text{den}} = a_{\text{den}}/(1 - a_{\text{den}})$, as it is well known, can be reliably measured, in fact, at values around 1 (i.e., log $Q_{\text{den}} = 0$). The displacement produced by changing pH keeps the curvature constant because the constant curvature amplitude is inversely proportional to the constant $\Delta C_p$.

Table 8. Values of Denaturation Entropy $S_{\text{den}}$ at Different Temperatures and Different pHs, for Lysozyme (from Pfeil and Privalov).

| $\ln T$ | $S_{\text{den}}$ (J K$^{-1}$), pH 7 | $S_{\text{den}}$ (J K$^{-1}$), pH 6 | $S_{\text{den}}$ (J K$^{-1}$), pH 5 | $S_{\text{den}}$ (J K$^{-1}$), pH 4 | $S_{\text{den}}$ (J K$^{-1}$), pH 3 | $S_{\text{den}}$ (J K$^{-1}$), pH 2 |
|--------|-------------------------------|-------------------------------|-------------------------------|-------------------------------|-------------------------------|-------------------------------|
| 5.610  | 10.46                         | 10.46                         | 21.34                         | 57.74                         | 121.75                        | 121.75                        |
| 5.646  | 247.27                        | 247.27                        | 258.15                        | 294.55                        | 358.57                        | 358.57                        |
| 5.681  | 476.14                        | 476.14                        | 476.14                        | 523.42                        | 587.01                        | 587.01                        |
| 5.698  | 587.43                        | 587.43                        | 598.31                        | 744.33                        | 807.93                        | 807.93                        |
| 5.747  | 910.86                        | 910.86                        | 921.74                        | 958.14                        | 1022.15                       | 1022.15                       |
| 5.778  | 1117.97                       | 1117.97                       | 1128.84                       | 1165.24                       | 1229.26                       | 1229.26                       |
| 5.809  | 1318.80                       | 1318.80                       | 1329.68                       | 1366.08                       | 1429.67                       | 1429.67                       |
| 5.838  | 1513.77                       | 1513.77                       | 1524.65                       | 1561.05                       | 1624.65                       | 1624.65                       |
| 5.867  | 1702.89                       | 1702.89                       | 1713.77                       | 1750.17                       | 1813.76                       | 1813.76                       |
| 5.895  | 1886.98                       | 1886.98                       | 1897.86                       | 1934.26                       | 1997.86                       | 1997.86                       |
| 5.922  | 2066.06                       | 2066.06                       | 2076.94                       | 2111.25                       | 2176.25                       | 2176.25                       |
These authors have reported a list of values of enthalpy and entropy for native (H_N and S_N) and denatured (H_D and S_D) lysozyme. The thermodynamic functions have been measured at six different values of pH (7−2). From the tabulated values, we have calculated the functions H_{den} = H_T - H_N and S_{den} = S_T - S_N (we recall that H_{den} and S_{den} are the observed experimental thermodynamic functions analogous to H_{dual} and S_{dual}, respectively). The calculated values of H_{den} are reported in Table 7, and the calculated values of S_{den} are reported in Table 8. The observed values of enthalpy and entropy as functions of T and of ln T, respectively, are shown in Figure 9A,B. Even the data reported by Pfeil and Privalov, therefore, confirm all of the properties typical of the ergodic algorithmic model (EAM) as found in any denaturation diagram. Both convoluted binding functions R ln K_{dual} = \{−ΔG_{dual}/T\} = \{(1/T)^*g(T)\} and RT ln K_{dual} = \{(T)^*g(ln T)\} can be derived, suited to describe coherently all of the properties of this important series of reactions. These results have been obtained as a development of the suggestion put forward by Lumry of considering the thermodynamic functions enthalpy and entropy as composed of two parts, thermal and motive, respectively. By examining the theoretical foundations of this proposal, we have concluded that the system of every hydrophobic hydration process is biphasic and that the dual-structure partition function {DS-PF}:

\[
\begin{align*}
\{\exp((\Delta H_{\text{mop}}/RT)\exp(\Delta S_{\text{mop}}/R))
\cdot \exp(-\Delta H_{\text{th}}/RT)\exp(\Delta S_{\text{th}}/R))
\end{align*}
\]

\[
\begin{align*}
= K_{\text{dual}}
\end{align*}
\]

(117)

represents the probability state of these processes. The probability state has been demonstrated to be homologous with dilution (i.e., reciprocal concentration) and as such it is amenable of experimental determination. On the other hand, by passing to the logarithm of the partition function and as we move from probability space to thermodynamic space, whereby we can experimentally determine free energy, enthalpy, and entropy. The fundamental homology relationship between ideal dilution d_{id}, partition function Z_{id}, formation quotient Q_K, and formation constant K (or other equivalent potential function) has permitted to show how the mathematical expression of each function, partition function, formation constant, formation quotient, or chemical potential can be considered, at constant temperature, as a configuration (dilution) change of entropy density (dS)_T = (R dln d_{id}(A))/T and reported on the same abscissa axis in the geometrical plane where we can plot the vector representation of Gibbs equation

\[
-\Delta G^0/T = -\Delta H^0/T + \Delta S^0
\]

(118)

The vector representation is referred to the abscissa axis, where dilution and configuration change of entropy density (dS)_T = (R dln d_{id}(A))/T are reported, and to the ordinate axis whereby thermal change of entropy intensity (−ΔH^0/T) is reported. The vector geometry shows how there is a necessary perfect equality between thermal entropy intensity vector −ΔH^0/T and an entropy density component vector δS_{id}. The equality is necessary because both are legs of an isosceles right triangle. The equality

\[
-\Delta H^0/T = \Delta S_{id}
\]

(119)

is the mathematical formulation of the ergodic property of the chemical systems. The ergodic condition requires that dispersion of energy in time is equivalent to dispersion of energy in space. In terms of molecular processes, this means that thermal change of entropy intensity, produced by temperature and, therefore, by molecular velocity, equals the change of entropy density produced by dilution (see Part II, Section 2).

In any hydrophobic hydration process, the hydrophobic heat capacity, ΔC_{hydr} results to be a remarkable characteristic quantity. The hydrophobic heat capacity ΔC_{hydr} is a constant that behaves as a phase-transition entropy intensity change (ΔH_{hydr}/T), similar to the Trouton constant. The Trouton law states that the ratio ΔH_{evap}/T_{evap} = ΔS_{evap} is constant for many liquids, independent of temperature. By accepting as legal, being hydrophobic heat capacity ΔC_{hydr} actually constant, the extrapolation of ΔH_{dual} to T = 0 and of ΔS_{dual} to InT = 0, we have been able to calculate the function ΔH_{not} separated from ΔH_{th} as well as ΔS_{not} separated from ΔS_{th}, respectively. From the separate functions in the thermodynamic space, we have gone back to the probability space. Thus, we have obtained a dual-structure product partition function {DS-PF} valid for every hydrophobic hydration process.

The product partition function \{DS-PF\} of eq 113 is the product of a motive partition function multiplied by a thermal partition function.
\{DS-PF\} = \{MPF\} \cdot \{TPF\}

The thermal partition function \{TPF\} is referred to the solvent, whereas the motive partition function \{MPF\} is referred to the solute. The solvent is represented by a nonreacting molecule ensemble (NoRemE), whereas the solute is represented by a reacting mole ensemble (REME). The thermal functions \(\Delta H_{\text{sol}}/T\) and \(\Delta S_{\text{sol}}\) do not contribute to free energy because they compensate each other at any temperature, giving null thermal free energy \((-\Delta G_{\text{sol}}/T = -\Delta H_{\text{sol}}/T + \Delta S_{\text{sol}} = 0\). On the other hand, \{MPF\} is referred to the solute, yielding non-null motive free energy, \((-\Delta G_{\text{mot}}/T \neq 0\). The introduction of the Lambert thermal energy factor \(\text{THEF}\) \(\Phi = T^{-\langle G_{s\text{sol}} \rangle}\) associated with the concentration is the source of ergodicity of the thermodynamic systems, generating the thermal equivalent dilution (TED) principle.

From \{DS-PF\}, an ergodic algorithmic model can be developed consisting of a set of parabolic binding functions \(R \ln K_{\text{dual}} = \{f(T)\}^*g(T)\) and \(RT \ln K_{\text{dual}} = \{f(T)\}^*g\ln(T)\) (see Appendix C and Part II,11 Section 2). The binding functions of any hydrophobic hydration process are curved parabolic functions. The geometrical properties of the binding functions are representative of the characteristics of the thermodynamic properties of each hydrophobic hydration process. In fact, the tangents to the binding functions correspond to the observed thermodynamic enthalpy \(\Delta H_{\text{dual}}\) and entropy \(\Delta S_{\text{dual}}\) respectively: \(\Delta H_{\text{dual}}\) for each compound is a linear function of \(T\) with slope \(\Delta C_{p\text{hydr}}\) whereas \(\Delta S_{\text{dual}}\) is a linear function of \(\ln T\) with identical slope \(\Delta C_{p\text{hydr}}\). This same coefficient \(\Delta C_{p\text{hydr}}\) is inversely proportional to the constant curvature amplitude of both parabolic binding functions in every compound examined and is, therefore, a constant independent of temperature \(T\) for each compound. Moreover, the hydrophobic hydration heat capacity \(\Delta C_{p\text{hydr}}\) results to be constant, independent of temperature, for chemical conditions, too. In fact, we have shown that \(\Delta C_{p\text{hydr}} = \pm \xi \cdot C_p\) (where \(C_p = 75.36\ J\ \text{K}^{-1}\ \text{mol}^{-1}\) is the isobaric heat capacity of liquid water), depends on the stoichiometry of the reaction (phase transition) between water components \(W_1\) and \(W_2\). The coefficient \(\pm \xi\) is the power of the ligand \(\lambda_{\text{W}}\) in an association or dissociation constant, \(K = a_{\lambda\text{W}}/a_{\lambda\text{W}} = a_{\lambda\text{W}}\), where \(a_{\lambda\text{W}}\) and \(a_{\lambda\text{W}}\) indicate activity of the species. The positive coefficient \(+\xi\) is referred to a reaction of iceber formation (class A) with dissociation constant \(K_{\text{dual}}\) whereas the negative coefficient \(-\xi\) is referred to a reaction of iceber reduction (class B) with association constant \(K_{\text{dual}}\). As a function of a pseudostoichiometric coefficient \(\pm \xi\), \(\Delta C_{p\text{hydr}}\) remains the same as far as the reaction is the same. For the same reason, the coefficient \(\Delta C_{p\text{hydr}}\) is equal for both enthalpy \((\Delta H_{\text{dual}} = \Delta H_{\text{mot}} + g(T))\) and entropy \((\Delta S_{\text{dual}} = \Delta S_{\text{mot}} + g(\ln T))\) functions.

By a general survey of the literature concerning the hydrophobic hydration processes, we can conclude that the proposal of considering these systems as biphasic with the inherent adoption of the dual partition function represents a novelty and is promising of profitable results in this important field.

One more point of advancement of this paper is that, by applying the thermal factor \(\Phi = f_{\lambda T} \cdot T^{-\langle G_{s\text{sol}} \rangle}\) to the concentration parameter \(x_{\lambda\text{T}}\), we have opened the way to computer calculations of the ergodic properties of chemical solutions.

### APPENDIX A

Solution Entropy and the Second Law of Thermodynamics

The second law of thermodynamics deals with energy dispersal, on which the idea of entropy was founded. The entropy change can be either entropy intensity or entropy density, which are equivalent (ergodicity). They can be summed up or subtracted from each other, notwithstanding the different molecular mechanisms:

(a) Entropy intensity change is produced by a temperature change \(\delta \ln T\).

(b) Entropy density change is produced by a dilution change \(\delta d_{\text{mot}}\).

Therefore, entropy \(S\) being a function of two variables \(T\) and \(d_{\text{mot}}\) as shown in eq 7, the relationship can be formalized as

\[ S = f(T, d_{\text{mot}}) \]

and by differentiating, in explicit form, we can write for component \(A\)

\[ \delta S = (C_p\lambda A) \ln T + (R) \ln d_{\text{mot}}(A) \]

By considering that for component \(A\)

\[ C_p\lambda A \ln T = C_p\lambda A dT/T = \delta_{\text{mot}} Q_{pA}/T \]

where \(\delta_{\text{mot}} Q_{pA}\) is the reversibly exchanged heat, we obtain the correct expression of entropy in solution

\[ \delta S = \delta_{\text{mot}} Q_{pA}/T + R \ln d_{\text{mot}}(A) \]

with \((\delta_{\text{mot}} Q_{pA}/T) \geq 0\) and \((R \ln d_{\text{mot}}(A)) \geq 0\). The second law of thermodynamics puts to entropy the condition

\[ \delta S = \delta_{\text{mot}} Q_{pA}/T \geq 0 \]

This old expression, usually found in the texts for the characteristic conditions of change imposed by the second law of thermodynamics, considers only entropy intensity but ignores the existence of entropy density. The old expression, in fact, is adequate for thermal machines doing mechanical work or in general for processes involving only heat transfers. The definition of the limitations of entropy \((\geq 0)\) follows the second law of thermodynamics. We can recall the definition of Clausius of the second law of thermodynamics (1856): "Heat can never pass from colder to warmer body without any other change, connected therewith, occurring at the same time". The expression of eq A.5 is, however, inadequate for reacting chemical systems involving concentration changes. We can add, therefore, a further sentence, concerning entropy density changes, to the definition of Clausius, as a consequence of eq S and inherent limitations. The following are the results of the complete second law, for systems out of equilibrium:

(i) Heat can never pass, at constant dilution, from colder to warmer body without any other change, connected therewith, occurring at the same time and

(ii) Molecules can never pass, at constant temperature, from more diluted to more concentrated solutions without spending chemical or electrochemical work.

Both definitions are valid and correspond to the common sense that confirms that as heat tends to pass spontaneously from hot to cold bodies to disperse energy, the solute in a solution diffuses from concentrated to diluted solutions, thus
partition functions. The monocentric partition function gives origin to a set of mathematical relationships as shown in Table B1.

Table B1. Development of Constant $K_m$ (Monocentered)

| K_m | Probability Space | Thermodynamic Space |
|-----|------------------|---------------------|
| probability $\rightarrow$ | $K_m = \exp(-\Delta G^*/RT)$ | monocentered PF |
| thermodyn. function $\rightarrow$ | $\ln K_m = (-\Delta G^*/RT)$ | property (B.1) |
| idem $\rightarrow$ | $R \ln K_m = -\Delta G^*/T = -\Delta H^*/T + \Delta S^*$ | straight line (B.2) |
| idem $\rightarrow$ | $\partial (R \ln K_m)/\partial (1/T) = -\Delta H^*$ | slope $-\Delta H^*$ (B.3) |
| idem $\rightarrow$ | $RT \ln K_m = -\Delta G^* = -\Delta H^* + T\Delta S^*$ | straight line (B.4) |
| idem $\rightarrow$ | $\partial(RT \ln K_m)/\partial T = \Delta S^*$ | slope $\Delta S^*$ (B.5) |

Obtaining dispersion of the energy carried by molecules. Both processes are entropic effects, whereby the system is evolving toward a state of maximum entropy:

(i) $(dS)_d = (\delta_{\text{sol}} Q_d/T) \geq 0$ (entropy intensity change, at constant dilution) and

(ii) $(dS)_T = (R \ln d_{\text{dil}}/A) \geq 0$ (entropy density change, at constant temperature).

**APPENDIX B**

From Monocentric to Dual-Structure Partition Function
If we consider an equilibrium for a normal chemical reaction, the constant $K$ represents a probability ratio, homologous with partition functions. The monocentric partition function $K_m$ gives origin to a set of mathematical relationships as shown in Table B1.

On the other hand, the experimental constant $K_{dual}$ for a hydrophobic hydration process gives birth to a set of mathematical relationships (Table B2), which is different from that of a normal constant. The characteristic property of the experimental constant $K_{dual}$ is that the binding functions $R \ln K_{dual} = \{f(1/T)g(T)\}$ and $RT \ln K_{dual} = \{f(T)g(\ln T)\}$ are convoluted parabolic curves. The curvature amplitude, $\Delta C_{p,\text{hydr}}$ (heat capacity), is invariably constant in each compound, identical for both enthalpy and entropy.

The heat capacity $\Delta C_{p,\text{hydr}}$ depends on the number $\xi_w$ of water molecules $W_i$ involved in each reaction and on the molar heat capacity of liquid water $C_{p,m}$ ($\Delta C_{p,\text{hydr}} = \pm \xi_w C_{p,m}$ where $\xi_w$ is an absolute number and $C_{p,m} = 75.36$ J K$^{-1}$ mol$^{-1}$). The number $\xi_w$ is positive (+) for convex curves and negative (−) for concave curves. The sign (+) is for compounds of class A (those forming iceberg $W_{II}$ from water $W_I$ (solvent)), whereas the sign (−) holds for compounds of class B (those transforming iceberg $W_{II}$ into water $W_I$ (solvent)). Water $W_I$ constitutes the solvent, whereas iceberg $W_{II}$ belongs to the solute. Values of $\Delta H_{\text{mot}}$ and $\Delta S_{\text{mot}}$ are calculated from the curves of Table B2. For enthalpy, the value of $\Delta H_{\text{mot}}$ is calculated by extrapolation to $T = 0$ of the tangent

$$\Delta H_{\text{dual}} = \Delta H_{\text{mot}} \pm \Delta C_{p,\text{hydr}} T$$

(B.11)

whereas for entropy, $\Delta S_{\text{mot}}$ is calculated by extrapolation to $\ln T = 0$ of the tangent

$$\Delta S_{\text{dual}} = \Delta S_{\text{mot}} \pm \Delta C_{p,\text{hydr}} \ln T$$

(B.12)

At this point, the question arose: why do we find binding functions as straight lines in normal reactions and as parabolic curves in hydrophobic hydration processes? We found an answer by considering that for normal reactions, we are dealing with a monophasic system represented by a monocentric partition function $K_m$ (e.g., van’t Hoff equation from...
Table B4. Development of Constant $\zeta_{th}^{\text{idem}}$

| $\zeta_{th} = 1$ |
|-------------------|
| Probability Space |
| $\zeta_{th} = \exp(-\Delta G_{th}/RT) = 1$, $-\Delta G_{th} = 0$ |
| Thermodynamic Space |
| $\ln \zeta_{th} = -\Delta H_{th}/RT + \Delta S_{th}/R = 0$ property |
| $R \ln \zeta_{th} = -\Delta G_{th}/T = -\Delta H_{th}/T + \Delta S_{th} = 0$ =0 |
| $\delta(\ln \zeta_{th})/\delta(1/T) = 0$ =0 |
| $\delta(R\ln \zeta_{th})/\delta T = \Delta S_{th}$ =0 |

“Degree symbol omitted.

monocentric function), whereas in hydrophobic hydration processes, we have two phases, pure solvent and solute. Each phase is represented by a constant: constant $\zeta_{th} = 1$ for the solvent and constant $K_{mot}$ for the solute, respectively, thus obtaining a product constant $K_{dual}$

$$K_{dual} = K_{mot} \zeta_{th} (\zeta_{th} = 1) \quad (B.13)$$

representing a dual-structure partition function \{DS-PF\}. The constant $K_{mot}$ represents a monophasic distribution as does any normal constant $K_{nu}$ whereas the experimental dual constant $K_{dual}$ represents a biphasic distribution. If the solution under consideration is extremely diluted in water, as is the case of every hydrophobic hydration process, the solvent in excess is at constant composition, hence at constant potential. Under these conditions, the molecular frame (MF) approach\(^\text{27}\) can be applied. According to MF, the solvent is considered a continuum, for example, to vacuum in ideal gas or as an implicit solvent in computational procedures. The mathematical development (Table B3) of $K_{mot}$ is practically identical to that of a normal constant $K_{nu}$ (cf. Table B1), whereas the development of $\zeta_{th}$ ($\zeta_{th} = 1$) is practically null (cf. Table B4).

By comparing Table B4 with Table B1, we observe that $K_{mot}$ depends on temperature $T$ in the same way as a constant $K_{nu}$ for a normal reaction (as in van’t Hoff equation), with constant $\Delta H_{mot}$. In Table 4, we observe that $\zeta_{th}$ depends exclusively on temperature ($\Delta H_{th} = \Delta C_{p,\text{hyd}}/T$). In fact, as far as the solvent is concerned, we observe that the solvent constitutes a non-reacting system for which free energy $\Delta G_{th}/T$ is invariably null, as shown in the following equations for protein denaturation

$$\Delta G_{th}/T = 0 - \left((\Delta C_{p,\text{hyd}}/T)\{(T_d - T) + T \ln(T/T_d)\}\right)$$

$$= 0 - \left((1/T_d) \int_{T=0}^{T_d} \Delta C_{p,\text{hyd}} \, dT\right)$$

$$- \int_{T=1}^{T_d} \Delta C_{p,\text{hyd}} \ln T \, dT = 0 - \{(\Delta H_{th}/T_d) - (\Delta S_{th})\}$$

$$= 0 - 0 \quad (B.26)$$

where $T_d$ is the temperature in the middle of transformation. Please note that the two integrals should be almost equal ($\approx$) because of the small ineffective difference in the lower limits $T = 0$ and $\ln T = 0$, respectively, under the condition that $T_d > 273 K \gg 1 K$ so that

$$-\Delta G_{th}/T = -\Delta H_{th}/T + \Delta S_{th} = 0 \quad (B.27)$$

because

$$\Delta H_{th}/T \approx \Delta S_{th} \quad (B.28)$$

Therefore, we can conclude that the whole hydrophobic hydration system is composed of two phases (solvent and diluted solute) and consequently the partition function comes out to be a dual-structure partition function \{DS-PF\}

$$(\exp(-\Delta G_{dual}/RT)) \approx \{(\exp(-\Delta H_{mot}/RT)\exp(\Delta S_{mot}/R))\} \cdot \{(\exp(-\Delta H_{th}/RT)\exp(\Delta S_{th}/R))\} \quad (B.29)$$

# APPENDIX C

**Parabolic Binding Functions from \{DS-PF\}**

By developing the dual-structure partition function \{DS-PF\}, we obtain curved binding functions $R \ln K_{dual} = \{f(1/T)*g(T)\}$ and $RT \ln K_{dual} = \{f(T)*g(ln T)\}$, which are convoluted functions, as regular parabolas. The double dependence on $T$ in $\{f(1/T)*g(T)\}$ and in $\{f(T)*g(ln T)\}$ is a consequence of the dual structure of \{DS-PF\} = \{M-PF\} \cdot \{T-PF\}. The second function $g(T)$ in $\{f(1/T)*g(T)\}$ or $g(ln T)$ in $\{f(T)*g(ln T)\}$, respectively, derived from the thermal factor $\{T-PF\}$, is the origin of the parabolic curvature induced to $R \ln K_{mot} = f(1/T)$ or to $RT \ln K_{mot} = f(T)$, respectively, which are linear functions derived from the motive factor $\{M-PF\}$. It is very useful, therefore, to show how the properties of each hydrophobic hydration process correspond to the geometrical properties of the parabola function.

The general equation for a parabola is

$$y = ax^2 + bx + c \quad (C.1)$$

and for a parabola passing through the origin is

$$y = ax^2 \quad (C.2)$$

In these parabolas, the Focus $F$ has coordinates $F(0, 0.5/a)$. To calculate the curvature amplitude of a parabola, we search for the value $x$ giving $y = 0.5/a$

$$y = ax^2 = 0.5/a \quad (C.3)$$

thus obtaining

$$x^2 = 0.5 \cdot a^{-2} \quad (C.4)$$

and hence

$$x = 0.5^{1/2} \cdot a^{-1} \quad (C.5)$$

from which we calculate the curvature amplitude $C_{\text{ampl}}$ (represented by segment F-C in Figure 10)

$$C_{\text{ampl}} = 0.7071a^{-1} \quad (C.6)$$

Equation C.2 corresponds to the thermodynamic functions
In these parabolas, the focus has coordinates $F(0, 0.5/\Delta Cp,\text{hydr})$, the curvature amplitude is $C_{\text{ampl}} = 0.7071/(\Delta Cp,\text{hydr})$. The first derivative

$$(−\partial (\ln K_{\text{dual}})/(\partial (1/T)))_{\Delta S} = \Delta H_{\text{mot}} + \Delta C_{p,\text{hydr}}T$$

is plotted in a diagram with axes: $x(T), y(\Delta H)$: the function is a straight line with slope $\Delta C_{p,\text{hydr}}$. The second mixed derivative results to be

$$(−\partial^2 (\ln K_{\text{dual}})/(\partial (1/T) \partial T))_{\Delta S} = \Delta C_{p,\text{hydr}}$$

with $C_{\text{ampl}} = 0.7071/\Delta C_{p,\text{hydr}}$.

The differentials at denominator ($\partial (1/T) \partial T$) correspond to the abscissa axes of the two successive diagrams, thus showing how the binding function $y(\ln K_{\text{dual}}) = \{f(1/T)*g(T)\}$ is a double-convoluted function in $(1/T, T)$ space. The binding function $RT \ln K_{\text{dual}} = \{f(T)*g(\ln T)\}$ is referred to axes: $x(\ln T), y(\Delta S)$: the function is a straight line with slope $\Delta C_{p,\text{hydr}}$. The second mixed derivative results to be

$$(\partial^2 (RT \ln K_{\text{dual}})/(\partial T \partial \ln T))_{\Delta S} = \Delta C_{p,\text{hydr}}$$

with $C_{\text{ampl}} = 0.7071/(\Delta C_{p,\text{hydr}})$.

The differentials at denominator ($\partial T \partial \ln T$) correspond to the abscissa axes of the two successive diagrams, thus showing how the binding function $RT \ln K_{\text{dual}} = \{f(T)*g(\ln T)\}$ is a double-convoluted function in $(T, \ln T)$ space.

**Common Second Derivative.** The second mixed derivatives of the two curved binding functions are equal.
Both binding functions are parabolas with equal curvature amplitude \( C_{\text{impl}} = 0.7071/\Delta C_{\text{phyd}} \).

The second mixed derivatives in eq D.5 represents the connection between the two binding functions \( R \ln K_{\text{dual}} \) and \( RT \ln K_{\text{dual}} : \Delta C_{\text{phyd}} \) cannot be nonconstant because it represents the constant curvature amplitude of both binding functions.

**Axis Transformation.** The matrix product

\[
\{ T_0 \} \cdot \left[ R \ln K \right] = \left[ R \ln K \right]
\]  

(D.6)

represents the transformation from binding function \( R \ln K_{\text{dual}} = \{(1/T)^*g(T)\} \) to binding function \( RT \ln K_{\text{dual}} = \{f(T)^*g(\ln T)\} \). The matrix \( \{ T_0 \} \) operates an axis transformation from axis \((1/T)\) to axis \((\ln T)\), by multiplication factor \((\ln T)\) (Figure 13).

![Figure 13. Axis transformation from space \((1/T, T)\) to space \((\ln T, T)\).](image)

The matrices of the product in explicit format become

\[
\begin{bmatrix}
    1/T & 0 & 1 \\
    0 & 1 & \ln T \\
\end{bmatrix} = \begin{bmatrix}
    R \ln T \\
    0 \\
\end{bmatrix}
\]

The function transformation \( R \ln K_{\text{dual}} = \{(1/T)^*g(T)\} \rightarrow RT \ln K_{\text{dual}} = \{f(T)^*g(\ln T)\} \) is not a function multiplication, rather a geometrical transformation from space \((1/T, T)\) to space \((\ln T, T)\). The binding function \( R \ln K_{\text{dual}} \) is expressed in entropy units \((J \, K^{-1} \, mol^{-1})\); the binding function \( RT \ln K_{\text{dual}} \) is expressed in enthalpy units \((J \, mol^{-1})\).

**Function Convolutions.** In functional analysis, convolution is a mathematical operation on two functions, \( f \) and \( g \), to produce a third function \( C(f \ast g) \), which expresses how the shape of one function \( f \) is modified by the other function \( g \).

The function couples \( \{(1/T)^*g(T)\} \) and \( \{f(T)^*g(\ln T)\} \), at constant \( d_{\text{dil}(A)} = \{1/\chi_{\text{A}}\} \), represent function convolutions. The convoluted functions are generated by the dual structure of the partition function

\[
\{ \text{DS-PF} \} = \{ \text{MPF} \} \cdot \{ \text{T-PF} \}
\]

The modified \( f \) functions, i.e., the first function \( f(1/T) \) in the convolution \( \{(1/T)^*g(T)\} \), or the first function \( f(T) \) in the convolution \( \{f(T)^*g(\ln T)\} \), respectively, are generated by \( \{ \text{MPF} \} \) (motive partition function), whereas the modifying \( g \) functions, i.e., the second function \( g(T) \) in the convolution \( \{(1/T)^*g(T)\} \), or the second function \( g(\ln T) \) in the convolution \( \{f(T)^*g(\ln T)\} \), respectively, are generated by \( \{ \text{T-PF} \} \) (thermal partition function).

In hydrophobic hydration processes, the \( f \) functions \( R \ln K_{\text{mot}} = f(1/T) \) and \( RT \ln K_{\text{mot}} = f(T) \), those to be modified by functions \( g \), are linear functions (e.g. van’t Hoff equation), whereas the convoluted functions are curvilinear.

**LIST OF SYMBOLS**

- \( x_A \) concentration of \( A \) in molar fraction
- \( \tau_i^\text{m} \) squared mean sojourn time
- \( \tau_i^\text{th} \) cumulative squared sojourn time
- \( a_A \) activity of \( A \)
- \( a_{X_A} = f(\tau_i^\text{m}) \) ideal calculated dilution of \( A \)
- \( \Phi = f(\tau_i^\text{m}) \) reciprocal THEF, to be associated with ideal dilution
- \( d_{\text{dil}(A)} = \{1/\chi_{\text{A}}\} \) ideal calculated dilution of \( A \)
- \( k_B \) Boltzmann constant
- \( \{ \text{DS-PF} \} \), dual-structure partition function
- \( \{ \text{MPF} \} \), motive partition function
- \( \{ \text{T-PF} \} \), thermal partition function
- \( \{ \text{ACSPF} \} \), partition function for normal reaction, with equilibrium constant \( K_{\text{eq}} \)
- \( \{ \text{Kdual} \} \), experimental equilibrium constant corresponding to \( \{ \text{DS-PF} \} \)
- \( C_{\text{mot}} = \phi_{\text{mot}} \) product equilibrium constant with \( \phi_{\text{th}} = 1 \)
- \( \chi_0 \) monolayer constant for normal reaction
- \( Q_a \) equilibrium quotient
- \( R \ln K_{\text{dual}} \) binding convoluted function in entropy units
- \( RT \ln K_{\text{dual}} \) binding convoluted function in enthalpy units
- \( DMSGN \) dimethionine derivative of chymotrypsinogen A

**ACKNOWLEDGMENTS**

This work is dedicated to the memory of Professor Gian Paolo Chiusoli, Emeritus, the University of Parma, who had highly appreciated this model as a landmark in the study of hydrophobic hydration processes and to the memory of Professor Rufus W. Lumry, Emeritus, the University of Minnesota, who had foreseen the distinction between thermal and motive thermodynamic functions and underlined the relevance of the dual structure of liquid water for a correct analysis of equilibria in biofluids. The authors acknowledge the University of Parma for financial support.
\[ C_{\text{PV}} = 75.36 \, \text{J} \, \text{K}^{-1} \, \text{mol}^{-1}, \text{molar heat capacity for liquid water} \]

\[ \Delta_{f \text{PV}} = C_{\text{PV}} = 75.36 \, \text{J} \, \text{K}^{-1} \, \text{mol}^{-1}, \text{entropy change for phase transition in pure water} \]

\[ \Delta C_{\text{hydr}}, \text{heat capacity in hydrophobic hydration processes} \]

\[ C_{\text{hydr}} = \frac{0.7071}{\Delta C_{\text{hydr}}} \text{ curvature amplitude of para bola} \]

\[ \Delta S_{\text{hydr}} = \Delta C_{\text{hydr}}, \text{entropy change for A or B phase transition of water} \]

\[ \delta_{\text{PV}} \text{, heat reversibly exchanged, at const. pressure} \]

\[ \text{class A, hydr. hydration process with reaction (phase transition) } A\left( \xi_{\text{w}} W_{\text{I}} \rightarrow \xi_{\text{w}} W_{\text{II}} \right) \text{ ice berg} \]

\[ \text{class B, hydr. hydration process with reaction (phase transition) } B\left( \xi_{\text{w}} W_{\text{II}} \rightarrow \xi_{\text{w}} W_{\text{I}} \right) \text{ iceberg} \]

\[ W_{\text{I}} \text{, no. of water clusters WI changing phase in a hydrophobic hydration process} \]

\[ \epsilon_{w} = \left( \sqrt{2 \pi m_{\text{w}} k_{\text{B}} T} \right)^{1/2}, \text{energy} \]

\[ \eta_{\text{w}} = \text{solute} \]

\[ \text{TED, thermal equivalent dilution principle } \left( -R \ln \left( \frac{X}{x} \right) = n_{\text{PV}} \cdot d \right) \]

\[ \Delta H_{\text{hydr}} \text{, experimental enthalpy} \]

\[ \Delta H_{\text{hydr}} = \Delta H_{\text{mot}} + \Delta H_{\text{th}} = \Delta H_{\text{hydr}} \left( \xi_{\text{w}} c_{\text{w}} T \right) = \left( \frac{\xi_{\text{w}} c_{\text{w}} T}{\xi_{\text{w}} c_{\text{w}} T} \right) \]

\[ \Delta S_{\text{hydr}} \text{, experimental entropy} \]

\[ \Delta S_{\text{hydr}} = \Delta S_{\text{mot}} + \Delta S_{\text{th}} = \Delta S_{\text{hydr}} \left( \xi_{\text{w}} c_{\text{w}} T \right) = \left( \frac{\xi_{\text{w}} c_{\text{w}} T}{\xi_{\text{w}} c_{\text{w}} T} \right) \]

\[ \Delta H_{\text{th}}, \text{thermal enthalpy} \]

\[ \Delta H_{\text{th}} = \Delta C_{\text{hydr}} T \]

\[ dS_{\text{th}} = \text{differential change of thermal entropy} = \text{differential change of entropy intensity} \]

\[ \Delta S_{\text{th}} = \Delta C_{\text{hydr}} \ln T \]

\[ \Delta G_{\text{th}} = \text{free energy (null)} \]

\[ \text{of gases in liquid water} \]

\[ \text{See footnote c, for extrapolation to } T = 0 \text{ and to ln } T = 0 \]

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