New and Notable

Preferential Interactions: It's as Simple as 1, 2, 3

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In a paper appearing in this issue, Record and Anderson provide a simple model for preferential interaction coefficients. Preferential interactions describe the mutual interference of solutes in a solution and are of fundamental importance in understanding linked equilibria in the most general way. Using the notation of Scatchard and Stockmayer, the simplest macromolecular system of interest can be reduced to a three-component system containing water (component 1), a macromolecule (component 2), and a solute (component 3). At constant temperature and pressure, the chemical potentials, μs, of these components are related through the fundamental Gibbs-Duhem equation:

$$n_1 d\mu_1 + n_2 d\mu_2 + n_3 d\mu_3 = 0, \quad (1)$$

where n denotes the number of mols of each component. If the macromolecule is used as reference, Eq. 1 can be rewritten as

$$-d\mu_2 = X_1 d\mu_1 + X_3 d\mu_3, \quad (2)$$

where X_1 and X_3 denote the number of mols of water and solute per mol of macromolecule. The effect of the solute on the macromolecule is given by the response function $\Gamma_{3,2}$

$$-\frac{d\mu_2}{d\mu_3} = \Gamma_{3,2} = X_3 + X_1\frac{d\mu_1}{d\mu_3} = X_3 - X_1\Gamma_{3,1}. \quad (3)$$

The crucial consequence of Eq. 3 is that the coefficient $\Gamma_{3,2}$ depends not only on X_3, but also on the effect of the solute on water, $\Gamma_{3,1}$. Many biologically relevant interactions involve solutes whose concentration is vanishingly small compared with the molality of water, so that $\Gamma_{3,1} = 0$ for all practical purposes. Other important interactions, however, involve solutes at concentrations where the effect on water activity must be taken into account. Relevant examples are the effects of salts and denaturants on proteins and nucleic acids. In many such cases, the interaction of the solute with the macromolecule is to be cast in terms of the *preferential interaction* coefficient $\Gamma_{3,2}$, to indicate that the observed effect is due to accumulation or exclusion of the solute in and from the "domain" where the macromolecule operates, relative to the solvent. Because $\Gamma_{3,2}$ depends explicitly on $\Gamma_{3,1}$ (see Eq. 3), developing a model for preferential interaction coefficients is tantamount to developing a model for the effect of the solute on water. This is a rather nontrivial task in the general case.

Previous attempts to model preferential interactions have yielded to the temptation of associating the X values in Eq. 3 with specific *binding* interactions, thereby simplifying the problem. So did the classical Wyman theory of linked equilibria and its Tanford "correction" for preferential binding effects, the solvent-solute exchange models of Schellman and Timasheff, and the earlier Record-Anderson-Lohman treatment of salt effects. In these treatments, X_1 and X_3 represent the amounts of water and solute *bound* per mol of macromolecule. The model of Record and Anderson (1995), on the other hand, fully exploits the generality intrinsic to the Gibbs-Duhem equation. Using an earlier idea of Inoue and Timasheff, preferential interaction coefficients are cast in terms of the properties of two domains: a *local* domain containing components 1 and 3 in association with a single molecule of component 2, and a *bulk* domain containing components 1 and 3 only. The solution is assumed to be dilute enough in terms of component 2 that the various local domains are widely separated from each other through the bulk domain. The essence of the Record-Anderson treatment is embodied by the Gibbs-Duhem equation in the local domain (see Eq. 3), with the coefficient $\Gamma_{3,1}$ derived from the properties of the bulk domain. The treatment makes no assumption on the nature of the X values, which are quite appropriately defined as the amounts of solute and water *associated* with the macromolecule in the local domain, and preferentially *accumulated* in, or *excluded* from this domain relative to the bulk. In addition, the treatment extends to the case of a solute present as a charged component along with its co-ion, and the case where the macromolecule itself is charged. These cases are dealt with by properly defining chemical potentials for the charged species in Eq. 3 and taking into account electroneutrality. The result is a general description of preferential interactions that is nowhere weakened by ad hoc assumptions on the nature of the interactions involved. This is a valuable contribution to the field.

Much of the value of the two-domain model is that it offers a simple interpretation of nonideality effects induced by the presence of the macromolecule in solution and makes precise predictions on the extent and properties of preferential interactions that can be tested experimentally. A great charm of thermodynamics is the ability to describe reality in the most general way. An even greater charm is the ability to *predict* reality, when all the components responsible for a given effect are identified correctly and dealt with. One of the most challenging tasks of biological thermodynamics is to predict energetics from structure. The two-domain model is a small, but important, step in this direction. It may serve as a framework to arrive at quantitative predictions about macromolecular interactions involving charged and uncharged components and should be particularly useful in Monte Carlo studies of counterion association with DNA. The impact of the two-domain model, however, will extend to the treatment of salt

Received for publication 5 January 1995 and in final form 5 January 1995.

0006-3495/95/03/727/02 $2.00

effects in general, because it significantly improves earlier treatments of specific binding interactions. By correctly identifying all of the driving forces responsible for salt-induced effects in macromolecular systems, it would be possible to sort out the contributions due to specific and nonspecific binding interactions and reach a predictive understanding of either components. This is a timely and important issue.

Those who still naively think of salts as "inert spectators" of a game dominated by the macromolecule and its "ligands" should ponder the recent work on ribosomal RNA (Lu and Draper, 1994), protein-DNA interactions (Overman and Lohman, 1994), and enzymes like dialkylglycine decarboxylase (Hohenester et al., 1994), pyruvate kinase (Larson et al., 1994), and thrombin (Ayala and Di Cera, 1994). Monovalent cations binding in the millimolar range can influence dramatically the properties of proteins and nucleic acids. These interactions appear to be widespread in biology, and a thermodynamic description along the lines indicated by Record and Anderson (1995) will come in quite handy.

REFERENCES

Ayala, Y., and E. Di Cera. 1994. Molecular recognition by thrombin: role of the slow→fast transition, site-specific ion binding energetics and thermodynamic mapping of structural components. *J. Mol. Biol.* 235:733–746.

Hohenester, E., J. W. Keller, and J. N. Jansonius. 1994. An alkali metal ion size-dependent switch in the active site structure of dialkylglycine decarboxylase. *Biochemistry.* 33:13561–13570.

Larsen, T. M., L. T. Laughlin, H. M. Holden, I. Rayment, and G. H. Reed. 1994. Structure of rabbit muscle pyruvate kinase complexed with Mn^{2+}, K^{+}, and pyruvate. *Biochemistry.* 33:6301–6309.

Lu, M., and D. E. Draper. 1994. Bases defining an ammonium and magnesium ion-dependent tertiary structure within the large subunit ribosomal RNA. *J. Mol. Biol.* 244:572–585.

Overman, L. B., and T. M. Lohman. 1994. Linkage of pH, anion and cation effects in protein-nucleic acid equilibria. *Escherichia coli* SSB protein-single stranded nucleic acid interactions. *J. Mol. Biol.* 236:165–178.

Record, M. T., Jr., and C. F. Anderson. Interpretation of preferential interaction coefficients of nonelectrolytes and electrolyte ions in terms of a two domain model. 1995. *Biophys. J.* 68:786–794.