Supporting Information for

Temperature dependence of local salt accumulation is sequence specific

Liang Ma[1] and Qiang Cui[1,2,*]

1: Graduate program in Biophysics, 2: Department of Chemistry and Theoretical Chemistry Institute, University of Wisconsin-Madison, WI, 53706

E-mail: cui@chem.wisc.edu

1. Convergence plots of ion accumulation for all simulations.

The convergence of ion accumulation around peptide surface is investigated by checking the convergence of N(r). N(r) is calculated for different lengths of MD trajectories for all simulations. N(r) is regarded converged if including additional simulation configurations doesn't change the averaged N(r). For all cases, after around 16 ns, N(r) is essentially converged.

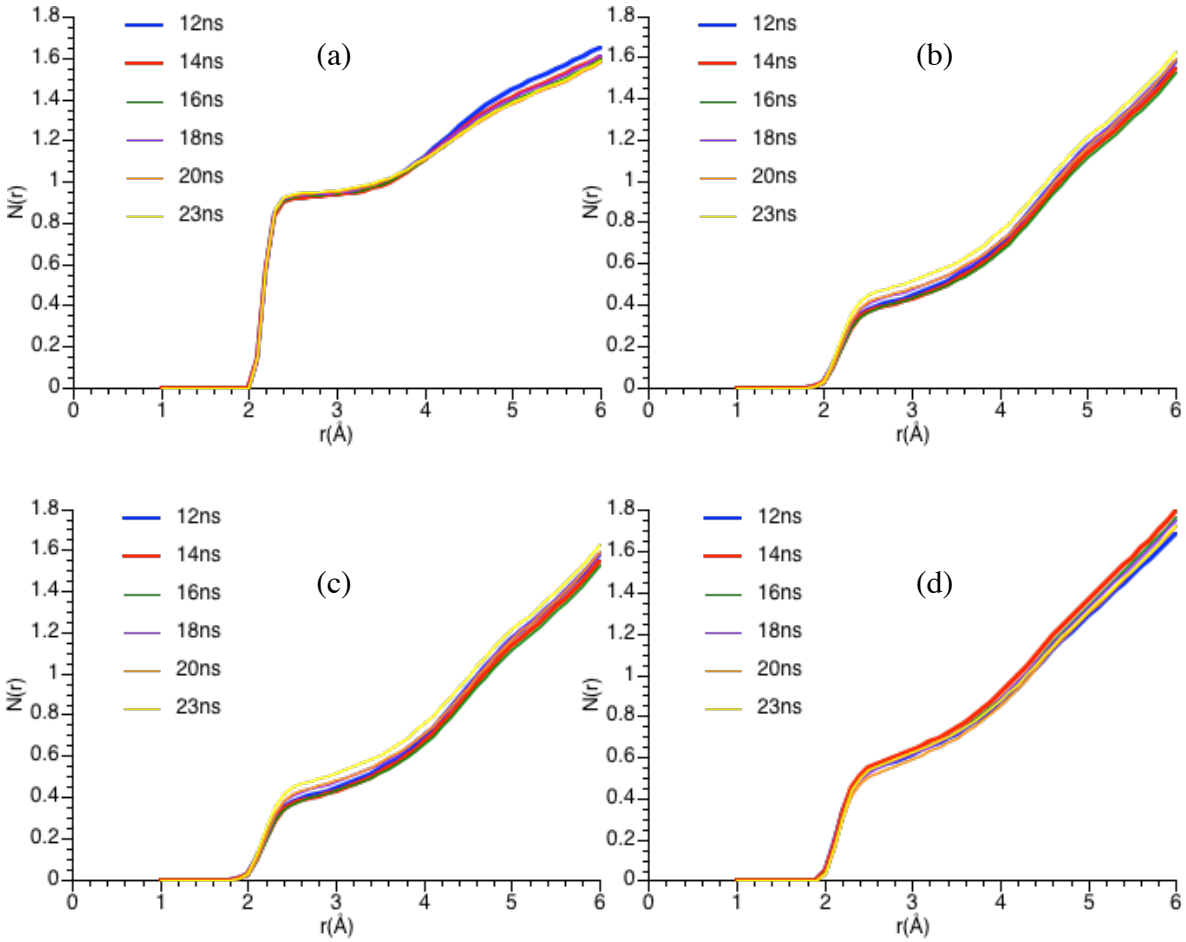


Figure S1. N(r) calculated from different lengths of MD trajectories for wild type simulations. (a) sodium from 10ºC simulation (b) chloride from 45ºC simulation (c) sodium from 10ºC simulation (d) chloride from 45ºC simulation. The label "12ns" refers to N(r) calculated from the first 12ns trajectory of the simulation, etc.

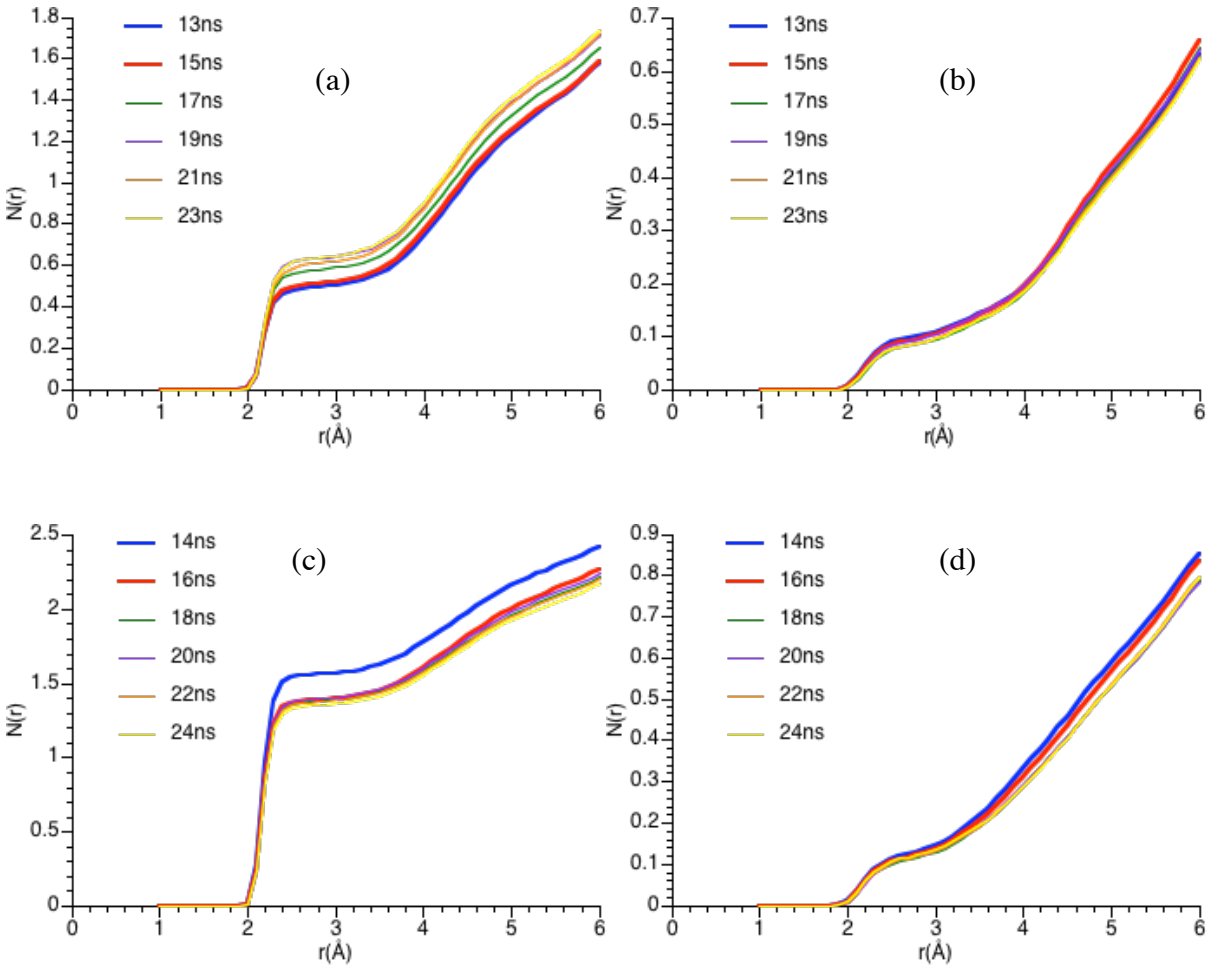


Figure S2. N(r) calculated from different lengths of MD trajectories for mutant simulations. (a) sodium from 10ºC simulation (b) chloride from 45ºC simulation (c) sodium from 10ºC simulation (d) chloride from 45ºC simulation. The label "13ns" refers to N(r) calculated from the first 13ns trajectory of the simulation, etc.

2. The association between sodium and chloride ions

To validate our discussion about salt-protein association, two equilibrium processes related to the salt-protein association process and their temperature dependence are also examined. These two processes include the ionic association between sodium and chloride ions and the sidechain association between positively charged sidechains and negatively charged sidechains, i.e. transient salt bridges (see Fig. 4 in the main text).

The radial distribution function g(r) and its integration N(r) of chloride ions around sodium ions at different temperatures are calculated for both the wild type and mutant peptide and the results are shown in Figure S3. The association between sodium and chloride ions becomes stronger at the higher temperature in both systems, which is expected since the association entropy for opposite charged species is positive. Importantly, the same trend in the sodium-chloride association for both peptides suggests that this process doesn't contribute significantly to the temperature dependence of ion-peptide association in the current systems.

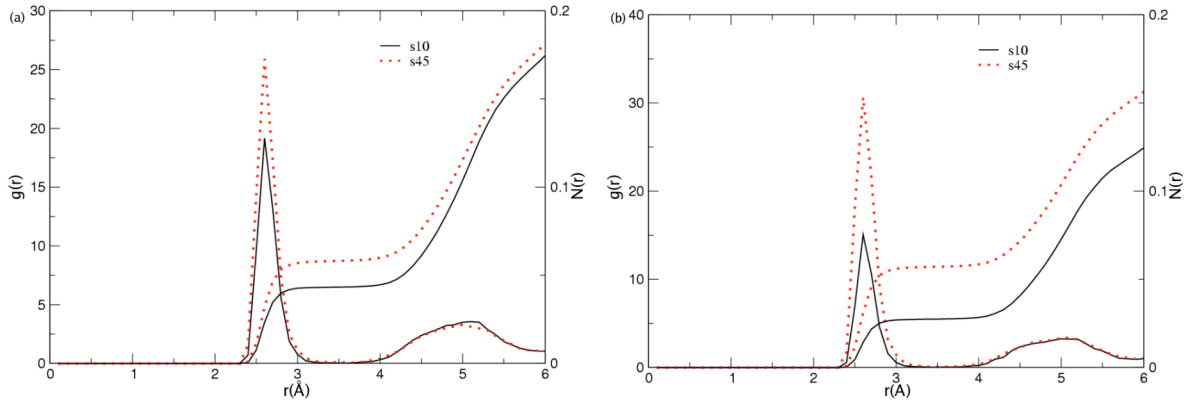
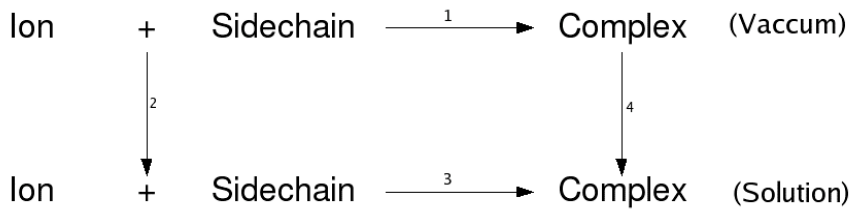


Figure S3. The radial distribution function g(r) and its integration N(r) of chloride ions around sodium ions from the simulation trajectories of (a) the wild type peptide and (b)

the mutant peptide. Label s10 and solid line refers to 200mM NaCl at 10°C and Label s45 and dotted line refer to 200mM NaCl at 45°C, respectively.

3. The binding free energy between ions and charged sidechains and its temperature dependence by an implicit solvation model based on Poisson-Boltzman/Solvent-Accessible-Surface-Area (PB/SASA) calculations.

To illustrate that the failure of PB to capture the interesting temperature dependence of sodium-peptide association (Fig. 5 in the main text) is largely due to the implicit treatment of ions, we consider the binding free energy between an explicit ion and an amino acid sidechain based on PB calculations. As discussed extensively in the literature, the binding free energy between an ion and a charged sidechain in solution can be calculated according to the thermodynamic cycle:



The binding free energy in solution corresponds to the free energy change for the step 3 can be calculated by

$$\Delta G_{bind} = -\Delta G_2 + \Delta G_1 + \Delta G_4 \qquad (1)$$

where ΔG_i represents the free energy change for the *i*-th step.

ΔG_1 represents the free energy change upon complex formation in vaccum and

$$\Delta G_1 = \Delta H_{vdw} + \Delta H_{coul} - T\Delta S_{vac}, \qquad (2)$$

where ΔH_{vdw} and ΔH_{coul} correspond the van der Waals and Coulombic interaction energy changes upon complex formation. The entropic contribution in vacuum, $T\Delta S_{vac}$, comes largely from the lost of translation/rotation upon association and is neglected in

the following discussions because the internal structure and the orientation of the sidechain are fixed in the calculations.

ΔG_2 corresponds to the hydration energy of ion and sidechain and

$$\Delta G_2 = \Delta G_{hyd}(Ion) + \Delta G_{hyd}(Sidechain) \quad (3)$$

ΔG_2 corresponds to the hydration energy of complex and $\Delta G_2 = \Delta G_{hyd}(Complex)$.

Therfore,

$$\Delta G_{bind} = \Delta H_{vdw} + \Delta H_{coul} + \Delta G_{hyd}(Complex) - \Delta G_{hyd}(Ion) - \Delta G_{hyd}(Sidechain) \quad (4)$$

(a) Calculation of ΔH_{vdw} and ΔH_{coul}

ΔH_{vdw} and ΔH_{coul} are calculated based on the CHARMM 22 force filed(*2*) and are taken as temperature independent.

(b) Calculation of ΔG_{hyd} for ion, sidechain and complex.

ΔG_{hyd} at certain temperature T is calculated based on the expression developed by Elcock and McCammon,(*2*)

$$\Delta G_{hyd}(T) = \gamma_{aliphatic}(T) \times SASA_{aliphatic} + \gamma_{aromatic}(T) \times SASA_{aromatic} + \gamma_{polar}(T) \times SASA_{polar} + \Delta G_{elec}(T) + 0.92 kcal/mol \quad (5)$$

where SASA is the solvent accessible area and can be divided into aliphatic, aromatic and polar, three regions and γ s represent proportionality constants which are different for aliphatic, aromatic and polar groups, and vary with temperature. $\Delta G_{elec}(T)$ is the electrostatic free energy change for transferring the ion, sidechain, or complex from vaccum to the dielectric continuum with a dielectric constant equal to that of water at the certain temperature.

The SASA is calculated using COOR SURF module in CHARMM program(*3*) with the radii set from the PARSE parameter set(*4*) and a solvent probe radius of 1.4 Å. γ_{polar} value is set to 5.4 cal/mol per Å^2 and temperature independent whereas $\gamma_{aliphatic}$, $\gamma_{aromatic}$ are obtained based on the equations by Elcock and McCammon:(*2*)

$$\gamma_{aliphatic} = 3.134694 + 0.1001063 \times T(^{\circ}C) - 4.418417 \times 10^{-4} \times T(^{\circ}C)^2 \quad (6)$$

$$\gamma_{aromatic} = 4.511007 + 0.0407585 \times T(^{\circ}C) - 2.928069 \times 10^{-4} \times T(^{\circ}C)^2 \quad (7)$$

$\Delta G_{elec}(T)$ is calculated by solving PB equations in vaccum and with a dielectric continuum and then taking the difference in the corresponding electrostatic free energies. The set of atomic radii established by Roux and co-workers(*5*) is used together with a solvent probe radius of 1.4 Å and a stern ion-exclusion of 2 Å. The partial charges are those in the standard CHARMM 22 force field(*1*) for proteins. The grid consists of 201^3 points spaced by 0.2 Å. The dielectric constant of the interior of the sidechain and that of vaccum is set to 1. The dielectric constant of water is taken as a function inversely dependent on the temperature; specifically, the value is 83.9, 71.6 and 55.1 at 10ºC, 45ºC and 85ºC, respectively. Once the electrostatic potential throughout the grid space is calculated, the electrostatic free energy of the system is obtained by:

$$G_{elec} = 1/2 \sum q_i \phi_i \quad (8)$$

The ions are placed on the collinear line of the sidechain at different differences from the sidechain, and the binding free energies are calculated according to Eq. 4. The binding free energies of four ion-sidechain pairs are shown in Figure S4.

Although the temperature dependence is small, it is important to note that different temperature dependences between different charge pairs are observed, as

expected based on the simple continuum electrostatic consideration discussed in the main text (Eq. 3 in the main text). This confirms that the failure of PB in capturing the interesting temperature dependence of ion-peptide association for the wild type peptide (Fig. 5 in the main text) is likely due to the implicit treatment of the ions.

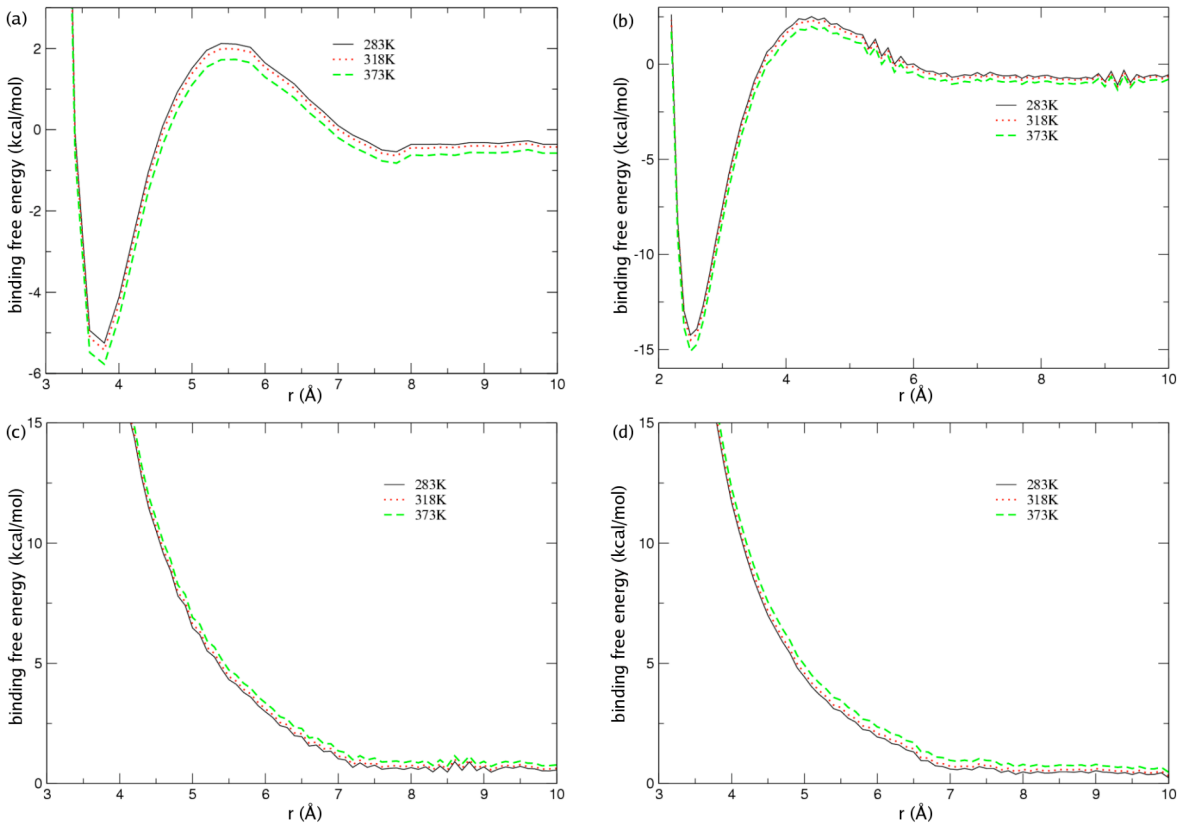


Figure S4. The binding free energy of four pairs of ion-sidechain by implicit solvation model at different temperatures 283K, 318K and 375K. (a) Arg and Chloride ion (b) Asp and Sodium ion (c) Arg and Sodium ion and (d) Asp and Chloride ion.

References

(1) MacKerell, A. D., Bashford, D., Bellott, M., Dunbrack, R. L., Evanseck, J. D., Field, M. J., Fischer, S., Gao, J., Guo, H., Ha, S., Joseph-McCarthy, D., Kuchnir, L., Kuczera, K., Lau, F. T. K., Mattos, C., Michnick, S., Ngo, T., Nguyen, D. T., Prodhom, B., Reiher, W. E., Roux, B., Schlenkrich, M., Smith, J. C., Stote, R., Straub, J., Watanabe, M., Wiorkiewicz-Kuczera, J., Yin, D., and Karplus, M. (1998) All-atom empirical potential for molecular modeling and dynamics studies of proteins. *J. Phys. Chem. B 102*, 3586-3616.

(2) Elcock, A. H., and McCammon, J. A. (1997) Continuum solvation model for studying protein hydration thermodynamics at high temperatures. *J. Phys. Chem. B 101*, 9624-9634.

(3) Brooks, B. R., Bruccoleri, R. E., Olafson, B. D., States, D. J., Swaminathan, S., and Karplus, M. (1983) Charmm - a Program for Macromolecular Energy, Minimization, and Dynamics Calculations. *J. Comput. Chem. 4*, 187-217.

(4) Sitkoff, D., Sharp, K. A., and Honig, B. (1994) Accurate Calculation of Hydration Free-Energies Using Macroscopic Solvent Models. *J. Phys. Chem. 98*, 1978-1988.

(5) Nina, M., Beglov, D., and Roux, B. (1997) Atomic radii for continuum electrostatics calculations based on molecular dynamics free energy simulations. *J. Phys. Chem. B 101*, 5239-5248.