Chinese Journal of Chemistry, **2007**, *25*, 1090—1093 **Full Paper**

Molecular Dynamics Simulation of the Binding Interaction between Hormone Glucagon Protein and Self-Assembled Monolayer Molecules

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Restrained molecular dynamics simulations were performed to study the binding affinity of the peptide with alkanethiols of different tail-groups, $S(CH_2)_7CH_3$, $S(CH_2)_7OH$ and $S(CH_2)_7COOH$, which self-assembled on Au(111) surface in the presence of water molecules. The curves of binding affinity were calculated by fixing the center of mass of the peptide at various distances from the assembling surface. Simulation results show that the binding affinity is in the order as COOH-SAMs＞OH-SAMs＞CH_3-SAMs, while 100% COOH-SAMs＞5% COOH-SAMs in concentration. The effects on binding affinity by different tail-groups were also studied. Results show that the binding affinity between COOH-SAMs and the peptide is bigger than those of the others and increasing the acidity of COOH-SAMs will result in stronger attractive power.

Keywords alkanethiol, binding affinity, molecular dynamics simulations, self-assembled monolayer

Introduction

Binding efficiency and selectivity of binding orientation are important factors in improving the detection of antibodies-antigens or protein microarrays. Self-assembled monolayers (SAMs) have been proposed as a platform for enhancing the detection by special designs with blockage of large spaces and randomness of the surface morphology. With reference to protein-surface interactions, much effort has gone into protein adsorption experiments and models over the past several decades[1-5] with ultimate aim to quantitatively measure, predict, and understand the detail of protein-surface interactions. As described by Norde[6] and Sigal,[7] proteins typically adsorb strongly to hydrophobic surfaces and weakly to neutral hydrophilic surfaces. Charged surfaces are generally found to be more adsorptive for oppositely charged proteins, and the degree of adsorption is typically lower for similarly charged surfaces and proteins.[6,7] While these trends are easily conceptualized, the numerous simulation interactions occurring among the functional groups of proteins, material surfaces, and surrounding body fluids are very complex in nature and the actual submolecular-level mechanisms and structural rearrangements involved in these reactions are not well understood.

Molecular simulation provides one of the most direct methods to theoretically investigate molecular behavior in complex systems, such as the adsorption of protein-surface systems. Because of the size of the molecular systems involved, the methods such as molecular dynamics (MD) and Monte Carol (MC), were required for simulation of these types of processes.[8] These methods employ potential energy (force-field) functions that calculate the overall potential energy of a system based on the summation of individual atom-atom pair interactions. The force field equations take into account the contributions from bonding interactions and nonbonding interactions. These energy contributions are determined by a set empirical parameters which were used to calculate energy by force field .

This study demonstrates the binding affinities between SAMs surfaces and a hormone glucagons protein.[9] The glucagon is a single α-helix basic protein which plays a major role in increasing blood glucose and maintaining normal concentrations of glucose in blood and is often described as having the opposite effect of insulin.[10]

The SAMs surfaces were individually generated with 1-heptanethiol derivatives [Au-$S(CH_2)_7$-X, X＝ COOH, OH and CH_3]. The change in different tail-groups will alter the binding affinity between protein

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Received October 8, 2006; revised January 11, 2007; accepted April 13, 2007.
Project supported by the National Center for High-performance Computing and the National Sun Yat-Sen University, Taiwan, China.

molecules and SAMs surfaces .

The present study performed molecular dynamics simulation on model systems of single glucagon protein on SAMs surfaces with carboxyl, hydroxyl and methyl tail-groups. Detailed analysis of the dynamics simulation reveals the physical mechanism of protein binding.

Molecular dynamics methods

Model systems of glucagons and SAM molecules

Several glucagon structures have been solved and deposited with protein data bank (PDB). We used the X-ray structure of glucagon (PDB ID: 1GCN) as the simulation model because the action and metabolism of glucagons[11] have been well studied.

The three kinds of SAM structures are 1-heptanethiol derivatives with different tail-groups (carboxyl, hydroxyl and methyl) and terminal thiol group bonding to gold atom, [Au-S(CH_2)$_7$-X] with X＝COOH, OH and CH_3. They were built and minimized by CHAMm program[12] with the additional force field shown in Table 1.[13,14]

Table 1 The additional force filed of Au and sulfur atoms

	Harmonic bond interaction	
	r_0/nm	k_r/(kJ•mol^{-1}•nm^{-2})
Au-S	0.02531	0.082
S-C	0.01836	0.085
	Harmonic angle interaction	
	θ_0	k_θ/(kJ•mol^{-1})
Au-S-C	109	193.84
	Dihedral angle interaction	
	φ_0	k_φ/(kJ•mol^{-1})
Au-S-C-C	180	1.29
S-C-C-C	−19	0.91
	Lennard-Jones interaction	
	r_{min}/nm	Epsilon/(kJ•mol^{-1})
Au	0.20736	0.32

Computational models and details

The molecular dynamics simulation was performed with NAMD[15] and the SAM-protein systems were studied by the MD software.

We used CHARMm force field and the additional parameters[13,14] for SAM-protein systems. The SAM surfaces were arranged in 16 by 16 hexagonal array with a gold-gold spacing of 0.475 nm[16] and 1-heptanethiol derivatives were tilted with 32°.[17] The COOH SAM surface was constructed with both COOH (protonated) and COO^- (depronated) functional groups. Five percents of the COOH groups were depronated and positioned throughout the SAM surface, as appropriate for a surface pK_a≈8.7[18] in a solution with pH＝7.4. And 100% of COO^- (depronated) functional groups of SAM surface were constructed. We moved the central of mass of protein on the surfaces of SAMs with 0.05, 0.1, 0.2 nm and generated TIP3P water boxes of volume 1.27 nm×0.47 nm×0.52 nm on the top of SAMs surfaces. The water molecules around the protein with 0.02 nm were deleted. The SAM-protein models are shown in Figure 1.

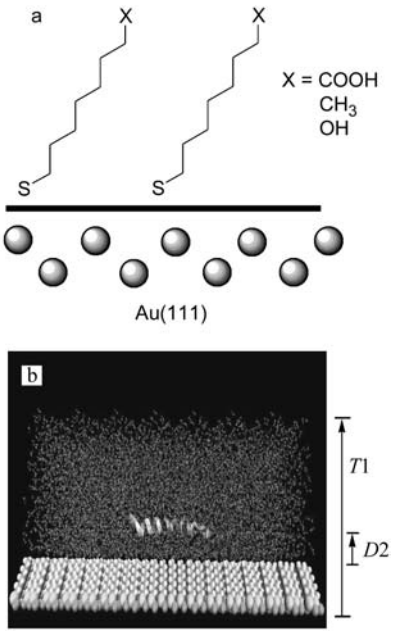

Figure 1 (a) The 1-heptanethiol derivatives with COOH, OH, CH_3 tail-groups are individualy coated on gold surfaces. (b) Snapshot of the glucagon-SAM surface in simulation. T1 : total height of simulation system (*T*1＝0.52 nm). *D*2: the separation distance between glucagon protein and SAM surface (*D*2＝0.05, 0.10, 0.15 nm).

The potential functions consist of bond, angle, dihedral, van der Waals (VDW), and Coulombic interactions. The full form of the potential functions is given by

$$U(r^N)=\sum_{\text{bonds}}\frac{K_l}{2}(l_i-l_{i,0})^2+\sum_{\text{angles}}\frac{K_\theta}{2}(\theta_i-\theta_{i,0})^2+\sum_{\text{torsions}}\frac{V_n}{2}(1+\cos[n\omega-\gamma])+\sum_{\text{VWD}}\varepsilon_{ij}\left[\left(\frac{A_{ij}}{R_{ij}}\right)^{12}-2\left(\frac{B_{ij}}{R_{ij}}\right)^{6}\right]+\sum_{\text{Coulombic}}\frac{332q_iq_j}{\varepsilon(r_{ij})r_{ij}}$$

where K_l, K_θ and V_n are the force constants for the bond, angle and torsion, respectively; while $l_{i,0}$, $\theta_{i,0}$ and γ are the values of bond length, bending angle and torsion angle at equilibrium. The Lennard-Jones 12-6 potential function was used to calculate VDW interactions, in which ε_{ij} and R_{ij} are the parameters for VWD depth and size, respectively. The last term is

Coulombic interactions in which q_i is the partial atomic charge.

The velocity Verlet algorithm was used for the integration of the equation of motion with time step of 2 fs. Initial velocities were assigned with Maxwell-Boltzmann distribution at 310 K and the simulation was performed at 310 K. During the initial simulation states, the harmonic constraints were used for SAM surfaces and glucagon proteins, and the bond length between hydrogen and any atom was fixed by SHAKE[10] algorithm. Since the contributions of the SAM-protein surface interactions come mainly from non-bonding interactions, we computed the electrostatic and van der Waals potential energy between SAM surfaces and protein molecules .

During simulation, we monitored the non-bonded interactions between SAM surface and protein molecules as a function of time. For SAM surfaces and glucagon proteins, the interaction curves become stable after 200 ps. After the simulation time (200 ps), we started to collect simulation data and the data collection took 400 ps for each simulation.

The binding energy was computed from non-bonding interactions between the SAM surface and protein molecule. The cutoff distance of non-bonding potential energy function is 1.2 nm.

Results and discussion

The hormone glucagon is a well-studied small all-α protein, which shows a characteristic topology (Figure 2). Figure 3 shows typical snapshots of the SAM surfaces composed of different tail-groups, such as carboxyl, hydroxyl and methyl tail-groups. Figure 4 and Table 2 show the binding affinity of glucagons protein to SAM surfaces with different separation distances (0.05, 0.1, and 0.2 nm). When the separation of glucagon-SAM surfaces was equal to 0.05 nm, the binding affinities of glucagon-SAM surfaces took the order: OH-SAMs＞COOH-SAMs＞CH_3-SAMs. With the separation distance of 0.1 nm , the binding affinities of glucagon-SAM surfaces were in the order of: COOH-SAMs＞OH-SAMs＞CH_3-SAMs. With the separation distance of 0.2 nm, the binding affinities of glucagon-SAM surfaces were close to zero. In the case of separation distance 0.05 nm, the Coulombic and VWD interactions (OH-SAMs-glucagon) were stronger than those (5% COOH-SAMs-glucagon). We considered 5% protonated carboxyl tail-groups of SAMs provided lower concentration of COO^- groups and caused the lower Coulombic interaction (SAMs-protein). Besides, we simulated the system involving 100% protonated COOH-SAM, glucagon protein, and water molecules. And the result is shown in Figure 5. From which it can be seen that the Coulombic interaction (100% COOH-SAMs-glucagon) is approximately 25 times of that (5% COOH-SAMs-glucagon). So, our results show that

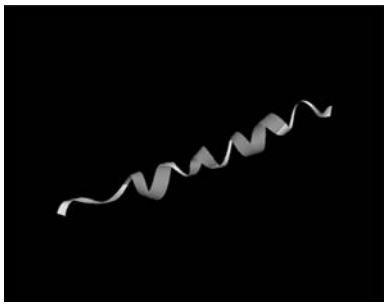

Figure 2 The structure of glucagon protein in a ribbon drawing.

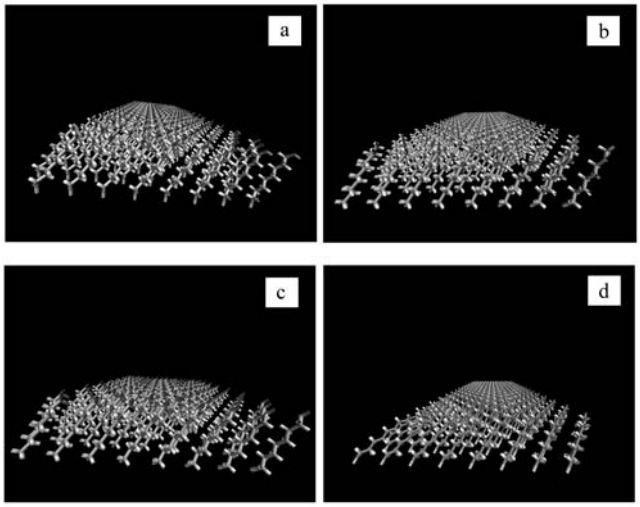

Figure 3 16×16 1-heptanethiol derivative molecules. (a) SAM surface with OH tail-groups. (b) SAM surface with CH_3 tail-groups. (c) SAM surface with 100% depronated COOH SAM molecules. (d) SAM surface with 5% depronated COOH SAM molecules.

Table 2 The binding affinities of glucagons-SAMs with different separation distance 0.05, 0.1, and 0.2 nm

SAMs	Affinity/ (kJ•mol^{-1})	Separation distance/nm		
		0.05	0.10	0.20
OH	Total	−534.92	−79.80	0
	Van der walls	−198.97	−33.99	0
	Coulombic	−335.95	−45.81	0
CH_3	Total	−117.41	−13.92	0
	Van der walls	−114.46	−13.79	0
	Coulombic	−2.95	−0.13	0
COOH[a]	Total	−243.56	−124.98	0
	Van der walls	−49.32	−25.70	0
	Coulombic	−194.24	−99.28	0
COOH[b]	Total	−4891.60	−3233.27	0
	Van der walls	−84.10	−18.85	0
	Coulombic	−4807.50	−3214.32	0

[a] COOH represents 5% depronated COOH SAM molecules. [b] COOH represents 100% depronated COOH SAM molecules.

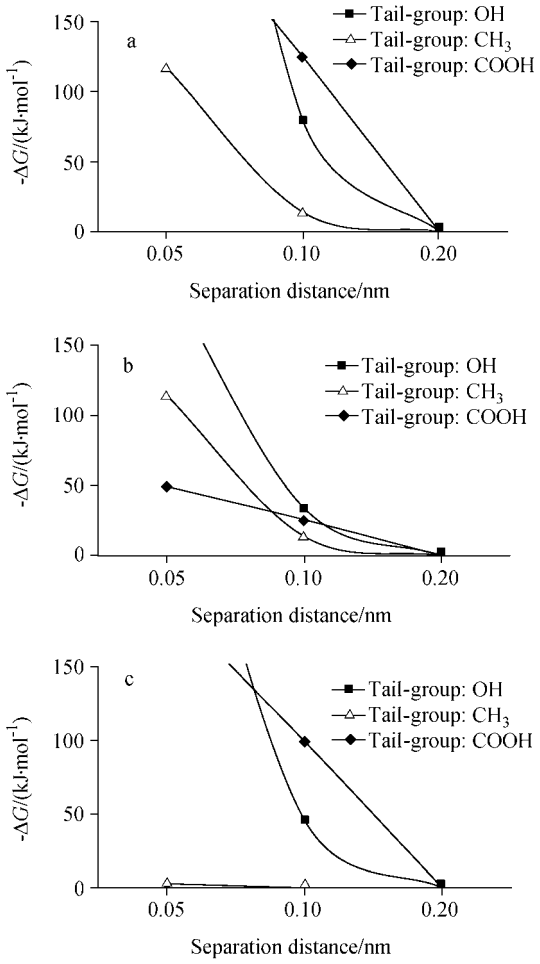

Figure 4 Comparing the binding energy between glucagon protein and SAM surfaces ([Au-S$(CH_2)_n$-X], X=COOH, OH and CH_3) (a) Coulombic+van der Waals energy profile, (b) van der Waals energy profile, (c) Coulombic energy profile.

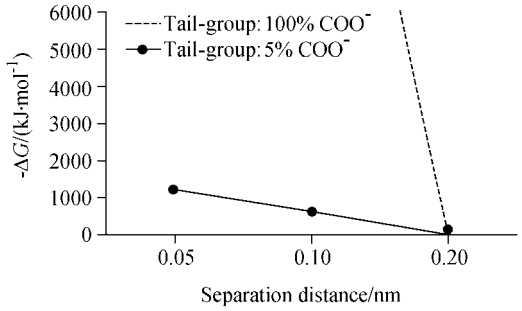

Figure 5 Comparing the binding energy of 5% and 100% depronated COOH SAM surface.

when the tail-groups of SAMs are more hydrophilic and carry negative electricity, the SAMs will show stronger attractive power to protein molecules.

Conclusion

In this paper, it was proposed that the use of different tail-groups of SAM surfaces may affect the binding affinity of protein molecules. The MD simulations of surface-protein interactions were carried out. In this work, the binding affinities between glucagon protein and three kinds of SAM surfaces were simulated. About the COOH SAM surfaces, the two kinds of COOH SAM surfaces (5% and 100% depronated COOH SAM molecules) were constructed. Our results show more hydrophilic and negatively electrical SAMs surfaces will possess stronger attractive power to proteins. In summary, molecular simulation is a valuable tool to predict the binding affinity of SAM-protein surfaces.

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(E0610087 LI, W. H.; DONG, H. Z.)