Adsorption and Reconfiguration of Amphiphiles at Silica-Water Interfaces: Role of Electrostatic Interactions, van der Waals Forces and Hydrogen Bonds

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ADSORPTION AND RECONFIGURATION OF AMPHIPHILES AT SILICA-WATER INTERFACES: ROLE OF ELECTROSTATIC INTERACTIONS, VAN DER WAALS FORCES AND HYDROGEN BONDS

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To my family
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ABSTRACT

The ability to explore and predict metastable structures of hybrid self-assemblies is of central importance for the next generation of advanced materials with novel properties. As compared to their thermodynamically stable forms, the kinetically stabilized materials show improved functionality potentially over their stable counterparts. The self-assembly processes usually originate from weak intermolecular interactions, involving a dynamic competition between attractive and repulsive interactions. These weak forces, including van der Waals (vdW), electrostatic interaction and the hydrogen bonding (H-bonding), can be tuned by external stimuli, e.g., confinement, temperature and ionization, and consequently driving hybrid materials into different configurations. It is challenging to determine the mechanisms and design rules for guiding a system into particular metastable states. Therefore, as a starting point, the focus of this dissertation is to understand how these interactions vary as a function of external parameters to determine why a particular equilibrium structure emerges. This information can not only be used to understand the mechanisms responsible for a system choosing a particular configuration, but also as a reference for future studies of metastable configurations with the long-term goal of developing rules for experimentalists to use in synthesizing hybrid materials into metastable states. In this dissertation, we studied the assembly processes of both ionic and non-ionic amphiphiles with silica that exhibit all three molecular interactions. By judiciously choosing three particular systems, the complex coupled nature of these interactions can be separated, allowing for each interaction to be isolated and systematically studied. For globular/silica hybrid system, the orientation of adsorbed protein depends on electrostatic interactions that can be tuned via ionization of silica; the morphologies of silica-adsorbed fatty acids are related to the competition between electrostatic and vdW interaction as a function of fatty acids’ ionized degree; the phase
behavior of ethoxylated surfactants in silica pores can be programmed by tuning the H-bonding as a function of temperature and confinement. For the first time, the atomic interactions at the amphiphile/silica interfaces are studied systematically to set up a basis for choosing the appropriate assembly environments to modulate the structures of a wide range of amphiphiles for wet bench research.
CHAPTER ONE
INTRODUCTION

Self-assembly in a hybrid material system is of interest due to its importance in diverse fields and applications, including material synthesis\textsuperscript{1–5}, catalysts\textsuperscript{6}, separation\textsuperscript{7}, energy production and storage\textsuperscript{8,9}, and biomedical applications\textsuperscript{10–12}. Hybrid self-assemblies can be prepared by the adsorption of soft materials on hard material surfaces or nanostructures. The adsorption process is initiated by the interaction between the hard adsorbents and the soft adsorbates, including hydrogen bonding and electrostatic interaction. These interactions depend not only on the surface chemistry of the hard adsorbent and soft adsorbates, but also on the external stimuli such as temperature, pH, and confinement. For decades, experimentalists have investigated the adsorption capacity and morphology of adsorbates as a function of various parameters, aiming to understand their effect on the assembly process with the goal of achieving the direct design of hybrid material systems guide with desired properties. However, what is lacking is an understanding of the mechanisms responsible for the observed structures at different experimental conditions even for systems in dynamic equilibrium. Thus, to uncover the mechanisms and to provide the experimentalists with the information on both the role of parameters on the observed structure but also the “rules” for adjusting/tuning these parameters to obtain a particular configuration, we conduct Molecular Dynamics (MD) simulations to understand how the atomistic interactions that are responsible for the bonding and overall structure, change as a function of external parameters.

In this dissertation, silica is chosen as the adsorbent, as the hydroxyl groups on silica not only provide the adsorption sites for hydrogen bonding but also enable surface modification for charge-charge binding. For example, the silica-based adsorbent can be negatively charged by ion exchange where the proton is replaced by a counterion. Furthermore, it can also be positively
charged by grafting organic functional groups such as alkylammonium to the surface. For the soft materials, we choose a wide range of amphiphilic molecules, including both ionic and non-ionic amphiphiles. Amphiphilic molecules consisting of hydrophilic and hydrophobic components are one of the most popular classes in soft matter. They have been widely studied for the past decades, offering a good reference point for direct comparison with our investigations. Most importantly, the differences in hydrophilic/hydrophobic groups for different types of amphiphiles allow us to study a wide range of molecular interactions under various parameters, that is by the judicious choice of a particular amphiphilic molecule, one key interaction can be studied thereby, providing a systematic approach for investigating these atomistic scale interactions and the effect on structure/property relationship.

For our studies, three hybrid material systems are chosen. The first is the globular protein/silica slab system, second, the fatty acid/silica slab system, and lastly, the ethoxylated surfactant/silica pore system. The orientation of the globular protein can be controlled by the electrostatic interaction between the protein and silica surface. This interaction can be varied by changing the degree of ionization of the silica surface, which leads to different equilibrium configurations. For the fatty acid/silica slab system, the aggregation of fatty acid monomers is driven by van der Waals interaction, while the adsorption of fatty acid assemblies is triggered by electrostatic interaction between the silica surface and fatty acids. We believe that the competition between those two interactions decides the morphology of the adsorbed fatty acid assembly, and the relative strength of these two competitive interactions can be tuned by varying the deprotonation state of fatty acids, leading to different adsorbed states of fatty acid aggregates. For the last system, the temperature-induced phase transition of ethoxylated surfactants in silica pores is due to H-bonds’ temperature responses that are different for surfactants with varying
hydrophilicity under different pore confinements. Thus, we focus on the H-bond interaction’s temperature responsiveness as a function of surfactants’ hydrophilicity and pore size, and its effects on the observed morphological transitions.

1.1 Dissertation Outline

The rest of this dissertation is organized as follows:

Chapter 2 covers fundamental concepts in statistical mechanics, molecular dynamic simulations, and algorithms used in this work.

Chapter 3 encompasses the study of the orientation of a globular protein (cytochrome c) adsorbed on a negatively charged silica surface. A detailed characterization of the orientation and the conformational stability for cytochrome c is provided, and equally important, the protein-silica electrostatic interaction as a function of the degree of ionization of silica is analyzed to uncover the mechanism responsible for the reorientation of the protein.

Chapter 4 describes the aggregation process of decanoic acids (C10 fatty acids) on a propylammonium functionalized silica surface. The effect of the degree of deprotonation of fatty acids is explored on the formation of disordered patches and ordered bilayers that are adsorbed on the silica slab. The silica-fatty acid electrostatic interaction and fatty acid-fatty-acid van der Waals interaction are analyzed as a function of the deprotonation degree of fatty acids, providing information on the correlation/competition between these interactions and the observed equilibrium structures. Furthermore, the equilibrium structures of silica-adsorbed fatty acid assemblies are compared to the fatty acid assemblies in bulk aqueous solution (without silica) to highlight the role of silica-fatty acid electrostatic interaction in inducing the bilayer structure that is essential in the application of protocells.
Chapter 5 focuses on the adsorption and aggregation process of ethoxylated surfactants (C₆E₃ and C₆E₅) as a function of temperature in a cylindrical silica pore (diameter of 8.6 nm). Both C₆E₃ and C₆E₅ show an aggregative adsorption behavior at an elevated temperature. This phenomenon is initiated by the temperature response of H-bonding, where the silica-water and surfactant-water H-bond interaction energy is decreasing as a function of temperature. The decrease of these H-bond interactions initiates the breakage of silica-water and surfactant-water hydrogen bonds, leaving both silica and surfactants dehydrated and leading to the aggregation of surfactants on the pore wall. Most importantly, the weaker C₆E₃-water H-bonding compared to the C₆E₅-water H-bonding leads to C₆E₃’s faster response to temperature in silica pores and higher thermal sensitivity. This mechanism provides insights into the tuning of the thermal stability of the ethoxylated surfactants in hydrophilic porous materials by changing the oxyethylene groups in surfactants.

Chapter 6 addresses the effect of pore size (diameter of 8.6 nm, 4 nm, and 2 nm) on the morphological transition and the temperature sensitivity of the ethoxylated surfactants (C₆E₅). We show that the decrease of surfactant-water H-bonding together with the increase of the van der Waals attraction between the assembly in and out of the pore is more hysteretic in a narrower pore, and that leads to the less sensitivity of the surfactant assemblies. Thus, the morphological transition temperature of the ethoxylated surfactants is shifted to a higher value in a narrower pore.

Chapter 7 summarizes the key observations, the correlated mechanisms responsible for the observations in our computational study, and how these mechanisms are related to experimental parameters for obtaining particular dynamic equilibrium configurations.
1.2 Background

This section provides the background knowledge for the materials used in this dissertation, including globular protein, saturated medium-chain fatty acids, ethoxylated surfactants, and silica. Their chemistry, phase behavior in aqueous solutions and the associated molecular interactions are reviewed, as shown in section 1.2.1, 1.2.2, 1.2.3, and 1.2.4, respectively. Specific problems regarding the molecular modeling for each material are also elucidated with the approach used to address these problems.

1.2.1 Immobilization of globular protein by tuning protein-solid surface interactions

A protein can be regarded as an assembly of amino acid groups, where the peptide-bonded amino acid groups with a specific sequence assemble and fold to a unique three-dimensional shape, also referred to as its native conformation\textsuperscript{13}. The globular protein is one of the common types, and spherical or ellipsoid-like can be referred to as “globular.” The globular shape of the protein is induced by its tertiary structure, where most hydrophobic amino acid side chains are buried and tightly packed in the interior of the protein, while the hydrophilic side chains are bound outwards, bringing them in contact with polar solvents (e.g., water). Consequently, globular proteins are relatively more soluble and stable in water than other types of protein, such as fibrous, disordered, and membrane proteins. It is important to recognize that a globular protein is considered as a sizeable amphiphilic molecule, making it surface-active similar to surfactants and fatty acids.

The adsorption and the immobilization of a protein on a solid surface have gained a lot of attention due to their broad range of nanotechnology applications, including biosensors\textsuperscript{14}, bioreactors, and bioanalytical devices. The performances of these devices can be highly influenced by conformational changes of the protein, and most often, in an unfavorable way. For example, in enzyme biosensors, if the catalytic site on the enzyme is not orientated to the target analyte, the
signal will be very low\textsuperscript{15}. Thus, to immobilize a protein in a preferred orientation is essential. This goal can be achieved by inducing and controlling the adsorption of protein on a solid surface with a preferred adsorbed state.

To better predict and manipulate the conformation of the adsorbed protein, the driving force that induces the adsorption needs to be recognized. It has been reported that the origin of interactions between protein and solid surface includes coulombic force, van der Waals forces, Lewis acid-base forces, and entropically based effects such as hydrophobic effect, conformational entropy, and restricted mobilities.\textsuperscript{16,17} Computational modeling and simulation of the protein on a solid surface can help researchers to recognize these interfacial interactions, to unravel the mechanisms that induce the binding of protein on a solid surface, and further to predict the behavior of complex protein systems in molecular details that cannot be directly measured in experiments.

As part of this dissertation, a particular globular protein, cytochrome $c$, will be investigated to determine its adsorbed orientation as a function of the degree of ionization of the silica surface and the binding mechanism in Chapter 3. It is proposed that the adsorption of cytochrome $c$ is induced by the electrostatic interaction between the protein and the silica surface, and the interaction can be further influenced by the degree of ionization of the silica surface. We will show that the change of the electrostatic interaction can lead to different adsorbed states and different orientations of the protein. Further, we will examine the conformational stability of the adsorbed protein with the existence of electrostatic attractions from the silica surface.

1.2.2 pH-responsive self-assembly of fatty acids

Fatty acids are crucial amphiphilic biomaterials due to their abundance in nature and their ability to self-assemble into vesicles in biomimetic applications\textsuperscript{18–20}. Fatty acids have also been suggested to be good models for pH-dependent targeted drug delivery owing to their pH-dependent
molecular formula and aggregation behavior\textsuperscript{21,22}. From a chemical point of view, a fatty acid molecule is an amphiphilic bio-carboxylic acid which has at least one hydrophilic carboxyl group (−C(= O)OH) connecting to a hydrophobic carbon chain that can be joined by single bonds, as in saturated fatty acids (C\textsubscript{n}H\textsubscript{2n+1}COOH), or by double bonds, as in unsaturated fatty acids (C\textsubscript{n}H\textsubscript{m}COOH). It can be at its protonated (RCOOH) or deprotonated (RCOO\textsuperscript{−}) state, depending on the acid/basic environment, characterized by the pH.

Figure 1.1. Titration curve for oleic acid/sodium oleate (C18). The regions for the formation of micelles, vesicles and oil droplets are demonstrated. The insets are the schematic representation of a fatty acid/soap bilayer fragment at intermediate pH and a soap micelle at high pH. The protonated headgroup of the fatty acid is represented by the black bead while the deprotonated headgroup is shown in white. This figure is reproduced from Ref.\textsuperscript{23} with the permission of the publisher.
One of the crucial properties characterizing the RCOOH/RCOO⁻ aqueous solution is $pK_a$ value, at which the number of fatty acid molecules at protonated states equals that at deprotonated states. The deprotonated/protonated states of fatty acids in aqueous solutions will directly influence their surface activity (ionization degree) and the aggregation behavior, and that can be programmed by adjusting the pH from below to above the apparent $pK_a$ value. As shown in Figure 1.1, it is reported that at pH $\approx pK_a$, the fatty acid soap (RCOO⁻ ⋯ HOOCR) is formed by ion-dipole interaction, and that facilitates the formation of bilayers at high concentration$^{23-25}$. However, at pH well above the apparent $pK_a$ where most fatty acids are present as deprotonated states, micelles can be formed$^{24,26-28}$. The bilayer-to-micelle morphological transition$^{27,29}$ at higher pH is believed to be induced by the increased electrostatic repulsion among headgroups of negatively charged fatty acids.

While most studies focus on fatty acids’ bulk self-assemblies, the aggregation behavior and phase state of fatty acids at solid-liquid interfaces are less known. As shown in Chapter 4, we systematically study the phase behavior and structure of medium-chain fatty acids on a silica-water interface as compared to their morphology in aqueous solution. We choose decanoic acid (C10 fatty acids) as the medium-chain fatty acid model and explore the effect of pH on the formation of disordered or ordered decanoic acid aggregates that are adsorbed on a charged silica surface. The vdw interaction among fatty acid alkyl chains and the electrostatic interaction between fatty acid and the silica surface will be analyzed to understand how the balance of these interactions leads to different structures as a function of pH. Given that the pH is a measure of the ratio of the free protons in the solution, the most widely used method to mimic the pH condition of a fatty acid solution in computational studies is to construct a mixture of fatty acid molecules and vary the ratio of the deprotonated monomers$^{24,30-32}$. We will also adopt this method in Chapter 4.
1.2.3 Temperature dependence of ethoxylated surfactants

An ethoxylated surfactant is one of the most conventional nonionic amphiphiles where a hydrophilic poly(ethylene oxide) chain is connected to a hydrophobic alkyl chain. It has a general structure of \( \text{H(\text{CH}_2)_n(\text{OCH}_2\text{CH}_2)_m} \), which can be abbreviated as \( \text{C}_n\text{E}_m \), enabling us to obtain surfactants with a wide variety of hydrophilic-lipophilic balance (HLB) by changing the \( n \) and \( m \) value. As shown in Figure 1.2, it is composed of an alkyl chain with \( n \) methylene groups and \( m \) hydrophilic oxyethylene units (EO group), and we choose two short-chain surfactants, \( \text{C}_6\text{E}_5 \) and \( \text{C}_6\text{E}_3 \), as representatives.

![Structural formula of alkyl polyglycol ether surfactants](image)

Figure 1.2. Structural formula of alkyl polyglycol ether surfactants (\( \text{C}_6\text{E}_5 \) and \( \text{C}_6\text{E}_3 \) as representatives) with indications of the hydrophobic tail and the hydrophilic head.

The self-assembly of surfactants towards the formation of micelles in the aqueous solution is thermodynamically driven by the competition between interfacial energy of the micelle core with water and the conformation distortion energy of the surfactant chains emanating from the core.\(^{3,33}\) In other words, it involves the hydration of hydrophilic head groups and the insertion of the hydrophobic tail in water, and that originates from weak molecular interactions such as hydrogen bonding, van der Waals interaction, electrostatic interaction, and hydrophobic effects.\(^{3,33}\) Specifically for ethoxylated surfactants in aqueous solution, hydrogen bonds (H-bonds) together with hydrophobic effect are essential for the formation of micelles. As shown in Figure 1.3, the hydrogen bond (\( \text{Dn-H} \cdots \text{Ac} \)) is an intermolecular interaction between a polar group \( \text{Dn-H} \) acting...
as the bond donor and a more electronegative atom acting as the bond acceptor.\textsuperscript{34,35} The polarity of OH group in the first water molecule and the electronegativity of O in the second water molecule forms an H-bond in water. In liquid water, the H-bond network can be formed as each water molecule is H-bonded with up to four other water molecules, as shown in Figure 1.3b. For a C\textsubscript{n}E\textsubscript{m} molecule in water, the hydroxide and oxyethylene groups of the C\textsubscript{n}E\textsubscript{m} molecule form H-bonds with water molecules, and the hydrophobic alkyl chain is inserted in water, leading to the solvation of the C\textsubscript{n}E\textsubscript{m} molecule, as illustrated in Figure 1.3c. As the concentration of C\textsubscript{n}E\textsubscript{m} is raised above its critical micelle concentration (CMC), more hydrophobic tails are inserted in water, and the H-bond network of water molecules is disrupted. The water molecules will be rearranged around the C\textsubscript{n}E\textsubscript{m} molecules and create cavities to accommodate the aggregates of alkyl chains.\textsuperscript{36,37} This process is known as the hydrophobic effect,\textsuperscript{37,38} leading to the aggregation of hydrophobic tails and the formation of the micelles in aqueous solution.

Figure 1.3. Schematic representations of (a) polarity of water molecules and the hydrogen bond in water, (b) hydrogen bond network of water, and (c) a C\textsubscript{6}E\textsubscript{3} molecule inserted in water. Carbon, oxygen and hydrogen are colored in grey, red and blue, respectively.). The H-bond is represented by a green dashed line. Water molecules in (b) are shown by the space-filling model, where the radii of the spheres are proportional to that of the atoms and the center-to-center distances are proportional to the distance between the atomic nuclei. The H-bond donor and bond acceptor are denoted in (a). In (c), the water molecules are rearranged around the C\textsubscript{6}E\textsubscript{3} molecule and the H-bond network is also reformed.
This mechanism decides that the phase transition of ethoxylated surfactant assemblies in aqueous solution is dependent on temperature, as the length and strength of H-bonds are sensitive to temperature\(^{39}\). As reported by Bock and Gubbins\(^{40}\), the surfactant-water H-bonding is weakened with increasing temperature. This reduction in H-bonding results in an increase in rotational entropy, i.e., the molecules rotate faster at higher temperatures, leading to the decrease of the surfactant’s solubility and phase separation above its *cloud point* \((T_{cp})\). Figure 1.4a is a phase diagram of the C\(_{12}\)E\(_8\)/water binary system. Along the temperature axis, the surfactants undergo the phase separation above \(T_{cp}\), where the micellar-rich phase and water-rich phase coexist, and the surfactant solution becomes cloudy. \(T_{cp}\) changes as a function of surfactant concentration, and the

![Figure 1.4](image)

**Figure 1.4. (a)** Phase diagram of C\(_{12}\)E\(_8\)/water binary system. Phase separation occurs as the temperature is increased above the cloud point.\(^{41}\) (b) Aggregation number of polyoxyethylene dodecyl ether with oxyethylene groups of 5, 6, 7, 8 as a function of temperature. Aggregation number of the micelle increases with temperature, and the surfactant with more oxyethylene groups is shown to be less sensitive to temperature. The inserted schematics show that the micelles have morphological transformation as a function of temperature before getting to the cloud point. Both figures are reproduced from Ref.\(^{41}\) with the permission of the publisher.
lowest one is defined as the *lower critical solution temperature (LCST)*. All ethoxylated surfactants, including \( C_6E_3 \) and \( C_6E_5 \) in this study, have this characteristic temperature-dependent behavior. Before reaching the cloudy point, the morphology of the micelles will undergo complex transformations. The size of the micelle can be characterized by measuring the aggregation number, which is defined as the number of surfactant monomers comprising one micelle. Nakama\(^{41}\) plotted the aggregation number of \( C_{12}E_5 \), \( C_{12}E_6 \), \( C_{12}E_7 \), and \( C_{12}E_8 \) micelles as a function of temperature, as shown in Figure 1.4b. The aggregation number of all these micelles increases with the temperature. Additionally, the surfactant with more oxyethylene groups is more hysteretic in response to temperature, indicating that it is less sensitive to temperature and more thermostable.

This H-bonding driven temperature-dependence characteristic of ethoxylated surfactants is advantageous in directing the adsorption and self-assembly of molecules in hydrophilic silica pores. Three types of H-bonds occur in the silica/\( C_nE_m \)/water system, namely (i) silica-water H-bonds, (ii) silica- \( C_nE_m \) H-bonds, and (iii) \( C_nE_m \)-water H-bonds. As part of this dissertation, we will show how these pairs of H-bond interaction energy change as a function of temperature, leading to the adsorption and aggregation of \( C_nE_m \) surfactants on the silica pore wall. In Molecular Dynamics simulation, the classical forcefield, including the ones we applied (see chapter 2), the H-bond interaction energy is treated as the nonbonded interaction, i.e., the sum of the electrostatic and vdW interaction. We will also elucidate the contribution of both interactions to the H-bonding. In Chapter 5, we focus on the temperature response of H-bonding for two surfactants with different oxyethylene groups, namely, \( C_6E_3 \) and \( C_6E_5 \). The aim is to understand the effect of surfactants’ hydrophilicity on the thermal response of H-bonding, leading to the different thermal stability of \( C_6E_3 \) and \( C_6E_5 \) in a silica pore. Further, in Chapter 6, we will explore the temperature response of
H-bonding in silica pores with different diameters, and we will show that the morphological transition temperature of the CₙEₘ assemblies can thus be tuned by changing the pore size.

1.2.4 Surface chemistry of silica materials

The interactions of the above-mentioned amphiphiles with solid materials are determined not only by the nature of amphiphiles but also by the surface chemistry of the solid surface. Silica is widely available in nature, and it is one of the most extensively used inorganic materials that utilize the molecular interactions with amphiphiles in applications like drug delivery, water purification, biosensors, and cosmetics. To perform our computational study on amphiphile-silica hybrid material systems, we need to understand the surface chemistry of silica and the existing silica model we can exploit.

The surface chemistry of silica nanoparticles of different sizes in aqueous solution varies greatly even in the same environment, i.e., the same pH, temperature, pressure, and ionic strength. Patwardhan et al.⁴² examined the sequences of silica-adsorbed peptides as a function of the diameters of silica nanoparticles (15-450 nm) at the same pH, and the peptide sequences show an 80% difference between that on small particles and large particles. The difference is due to the variance in the ionization degree and the silanol group density for silica particles with different sizes, which is referred to as the difference in Q², Q³, and Q⁴ environments⁴³,⁴⁴ on the silica surface. Here, the Q² surface is the silica surface terminated with two hydroxide groups per Si atom [(Si–O–)₂Si(–OH)₂], the Q³ surface is terminated with one hydroxide group per Si atom [(Si–O–)₃Si(–OH)], and the Q⁴ surface is with zero silanol group [(Si–O–)₄Si].⁴⁵ The silica surface can be ionized by replacing the proton (H atom) with counterion, M (M = Na, K et al.). The total density of SiO(H, M) groups per nm² on the surface varies from 0 to 9.4 /nm², depending on the synthesis recipe, the cleavage, the silica crystal type, and thermal treatment.⁴⁶–⁴⁹
For large ionic amphiphiles like peptides or protein, it is crucial to take surface ionization into account as the adsorption of ionic amphiphiles is primarily driven by ion-pairing (electrostatic interaction). It is only recently that simulations begin to take surface ionization into account for silica nanoparticles. The silica model database\textsuperscript{50} developed by Heinz’s group has shown to be able to predict the aqueous interfacial properties of all types of silica, which is substantiated by extensive comparisons to experimental measurements\textsuperscript{42}. Thus, we will utilize this database to explore the protein adsorption on the silica surface as a function of surface ionization, as shown in Chapter 3.

Apart from the bare silica, organic groups-functionalized silica, such alkylamine-functionalized silica has also attracted significant attention in catalysis, separation, and bio-related applications, owing to the stability of silica bulk and the flexibility of alkylamine terminal groups. Moreover, the silica surface terminated with alkylamine groups can be protonated to provide a positively charged surface that controls the adsorption or release of negatively charged amphiphiles, such as fatty acids and anionic drug molecules. We will also use this positively charged silica surface to investigate the structure and phase behavior of fatty acid assembly with different degrees of deprotonation. We will construct an α-quartz silica surface grafted with a uniform distribution of protonated alkylamine groups. The recipe for modeling an amine-functionalized silica surface can be found elsewhere as well as in Chapter 4.

Silica-based porous materials are also essential for their use as adsorbents or catalytic supports in the application of adsorption, filtration, and catalysis. Among porous materials, SBA-15\textsuperscript{51}, MCM-41\textsuperscript{52}, and MCM-48\textsuperscript{52} have obtained much attention due to their simple pore shape and pore connectivity compared to that of Controlled Pore Glass (CPG) and Vycor. The average pore diameter varies from 2 nm to 20 nm. In most theoretical and simulation studies, SBA-15 or MCM-
are modeled as pores with regular cylindrical geometry and fully hydroxylated.\textsuperscript{53–56} We will adopt the same treatment for silica pore to study the adsorption and self-assembly of nonionic ethoxylated surfactants in silica pores in Chapter 5 and Chapter 6.
CHAPTER TWO
MOLECULAR DYNAMICS SIMULATIONS

Molecular Dynamics (MD) is frequently used in the field of science and engineering including physics, chemistry, chemical engineering, mechanical engineering, etc. to understand the property of assemblies in terms of their structures and interactions. It serves as a bridge between the microscopic properties and the macroscopic scale variables in experiments such as temperature, pressure, volume, and viscosity of a liquid. MD provides a classical description of atoms/particles in a material system, including positions and velocities via Newton’s equation of motion and interaction energies between two particles through empirical potential parameters that are obtained by either fitting to experiment or first principles electronic structure calculations. MD can provide crucial mechanistic information concerning structure/property relationship of nanoscale configurations that are due to atomistic interactions.

In first principles electronic structure modeling, the movements and interactions of the electrons and atoms can be determined by the time-dependent Schrödinger or Dirac equation. However, one of the most efficient scaling methods with increasing system size are methods based on Density Functional Theory (DFT) typically scales as $O(N^3)$ (where $N$ is the size of the system), and the Higher Order Quantum Chemistry methods scales exponentially as $O(N^4)$, rendering the simulation of large systems untenable. On the other hand, in classical MD, the potential and forces are based on empirical formulations leading to $O(N)$ scaling with increasing system size, $N$. A typical MD simulation procedure is shown in Figure 2.1. To model the system of interest, the potential energy among all atoms is assumed to be pairwise additive and depend on special type coordinates. An initial configuration and an empirical potential appropriate for the given system are provided. The force acting on atom $i$ is calculated from the potential by using the central force theorem, $F_i = -\nabla_i U$. The position ($\mathbf{r}_i$) and velocity ($\mathbf{v}_i$) of each atom are updated at each timestep.
by integrating Newton’s equation of motion, $F_i = m_i \ddot{r}_i$ (or $F_i = m_i \dot{v}_i$). This procedure is repeated until the time-correlation function of microscopic properties converge to a constant value, indicating the equilibration of the system, and hence, a trajectory in phase space.

Figure 2.1. The flow chart of the Molecular Dynamics simulation procedure.
2.1 Force Calculation

The potential energy $U$ used to calculate the force, which is comprised of non-bonded and bonded interaction terms, is shown in equation 2.1. The non-bonded term describes vdW interaction and electrostatic interaction in our amphiphile/silica system, as described in section 2.1.1. The bonded interactions, including bond stretching, bond bending, and angle torsion, and will be elucidated in section 2.1.2.

$$U = U_{\text{non-bonded}} + U_{\text{bonded}}$$  \hspace{1cm} (2.1)

2.1.1 Non-bonded interactions

To describe the vdW potential, the 12-6 Lennard-Jones (LJ) potential\textsuperscript{[57–59]} will be employed (see equation 2.3). To account for the long-range electrostatic interaction, the Ewald procedure will be used (see equation 2.4) to accelerate the slowly convergent terms, such as charge-charge interaction.

$$U_{\text{non-bonded}} = U_{\text{LJ}} + U_{\text{Ewald}}$$  \hspace{1cm} (2.2)

Lennard-Jones (LJ) potential, $U_{\text{LJ}}$ is a simple pairwise potential that can model weak vdW interactions, it has the expression:

$$U_{\text{LJ}} = \sum_{ij} 4\varepsilon_{ij} \left[\left(\frac{\sigma_{ij}}{r_{ij}}\right)^{12} - \left(\frac{\sigma_{ij}}{r_{ij}}\right)^6\right]$$  \hspace{1cm} (2.3)

where $\varepsilon_{ij}$ is the depth of the potential well, a characteristic energy scale measuring how strong the two atoms attract each other; $\sigma_{ij}$ is a characteristic length scale measuring how close two nonbonded atoms can approach each other; $r_{ij}$ is the distance between two nonbonded atoms, $i$ and $j$. The $\frac{1}{r_{ij}^6}$ term describes the vdW attraction, and the $\frac{1}{r_{ij}^{12}}$ term is the hard-wall potential describing Pauli repulsion due to overlapping electron orbitals when two atoms come in close
contact\textsuperscript{60,61}. Thus, classically, the hard-wall potential prevents the atoms from coming too close to one another.

The LJ potential for one pair of nonbonded atoms as a function of interatomic separation is plotted and shown in Figure 2.2. The minimum of the potential (bottom of the potential well, $\nabla_i U = 0 = F_i$) corresponds to a distance between two nonbonded atoms, $r_m$, where $r_m = 2^{1/6} \sigma_{ij} \approx 1.122 \sigma_{ij}$. The force between each pair of nonbonded atoms can be determined from the central force theorem by taking the first derivative of the LJ potential. At the distance below $r_m$, the repulsive force (with a positive sign) dominates, pushing the atoms apart, while at a distance larger than $r_m$, the vdW attractive force (with negative sign) starts to dominate.

![Lennard-Jones potential graph](image)

**Figure 2.2.** Lennard-Jones potential energy between atom $i$ and atom $j$ as a function of their distance.

Since the atoms in the system can carry electrostatic charges, the Coulomb potential needs to be added to the simulated system. The Coulomb interaction, $\sum_{ij} f \frac{q_i q_j}{r_{ij}}$, decrease as a function of
\( r_{ij} \), which leads to a slowly non-uniformly convergent sum due to the long-range tail associated with the \( \frac{1}{r_{ij}} \) term. Ewald’s method is used to accelerate the convergence of this sum by effectively splitting this real-space sum into two parts, one in real-space and the other in reciprocal space along with a third term that cancels the \( i = j \) term (so-called self-interaction term) in \( U_{dir} \).

As shown in equation 2.4, \( U_{Ewald} \) has a constant term, \( U_0 \) and two quickly converging terms, \( U_{dir} \) and \( U_{rec} \). \( U_{dir} \) represents real space term containing a rapidly converging complementary error function, and \( U_{rec} \) represents reciprocal space (\( \vec{k} - \text{space} \)) term, which is a sum over a Gaussian type term that dampens the oscillatory term in the sum, resulting to a rapidly convergent summation (note, both terms contain Ewald parameter \( \beta \)). The system needs to be treated with periodic boundary conditions, due to the use of Fourier transform in Ewald’s method, which requires reciprocal space. For example, the cutoff in the real-space sum for a given \( \beta \) is typically on the order of 1 nm as complementary error function \( \text{erfc}(r_{ij,n}) \) converges quickly in the range of \( 0 < r_{ij,n} < 1 \) while decays slowly in the range of \( 1 < r_{ij,n} < 2 \).

\[
U_{Ewald} = U_{dir} + U_{rec} + U_0 \\
= \frac{f}{2} \sum_{i,j}^{N} \sum_{n_x} \sum_{n_y} \sum_{n_z} q_i q_j \frac{\text{erfc}(\beta r_{ij,n})}{r_{ij,n}} \\
+ \frac{f}{2\pi U_{Ewald}} \sum_{i,j}^{N} q_i q_j \sum_{m_x} \sum_{m_y} \sum_{m_z} q_i q_j \exp \left( -\left( \frac{\pi m}{\beta} \right)^2 + 2\pi i m \cdot (r_i - r_j) \right) \\
- \frac{f\beta}{\sqrt{\pi}} \sum_{i}^{N} q_i^2
\]  

(2.4)

In equation 2.4, \( q_i \) and \( q_j \) is the charge on atom \( i \) and \( j \), respectively; \( \epsilon_0 \) is the electric constant, \( 8.854 \times 10^{-12} \text{ F.m}^{-1} \); \( f = \frac{1}{4\pi \epsilon_0} \); \( n = (n_x, n_y, n_z) \) is the index vector of the simulation box; \( r_{ij,n} \)
is the distance between the charged atoms; $\text{erfc}(x)$ is the complementary error function. $\beta$ is Ewald’s parameter that determines the relative weight (number of terms) of the sums between real and reciprocal space, and $m = (m_x, m_y, m_z)$ is the wave vector in reciprocal space.

### 2.1.2 Bonded interactions

The bonded interactions, including bond stretching, bond bending, and angle torsional interaction potential are represented by a harmonic potential as shown in equation 2.6, 2.7 and 2.8, respectively. The geometry of a simplest molecule is illustrated in Figure 2.3, $i, j, k, l$ is in the sequence of covalently bonded atoms, $r_{ij}$ represents the distance between bonded atoms, $\theta_{ijk}$ is the angle between vector $r_{ij}$ and $r_{jk}$, and $\phi_{ijkl}$ is the angle between plane $ijk$ and plane $jkl$.

![Geometry of a molecule with bonded atoms](image)

Figure 2.3. Geometry of a molecule comprising atoms of type $i, j, k$ and $l$. The definition of distance $r_{ij}$, bend angle $\theta_{ijk}$ and torsion angle $\phi_{ijkl}$ is shown schematically.

\[
U_{\text{bonded}} = U_{\text{stretch}} + U_{\text{bend}} + U_{\text{torsion}}
\]  

\[
U_{\text{stretch}} = \sum_{ij} \frac{1}{2} k_{r,ij} (r_{ij} - r_{eq,ij})^2
\]

\[
U_{\text{bend}} = \sum_{ijk} \frac{1}{2} k_{\theta,ijk} (\theta_{ijk} - \theta_{eq,ijk})^2
\]
\[ U_{\text{torsion}} = \sum_{ijkl} \sum_{m} \frac{1}{2} k_{\phi m,ijkl}(1 + \cos(m\phi_{ijkl} - \phi_{eq,ijkl})) \]  

(2.8)

In these equations, \( r_{eq,ij} \) is the equilibrium bond length, \( \theta_{eq,ijk} \) and \( \phi_{eq,ijkl} \) is a phase shift angle where for \( m = 0 \) it acts as an equilibrium angle, and \( m \) is related to the symmetry of the dihedral angle that reflects the periodicity of the dihedral angle (how often it repeats). \( k_{r,ij}, k_{\theta,ijk} \) and \( k_{\phi m,ijkl} \) are the harmonic force constants. These parameters are typically determined by quantum mechanical calculations combined with thermophysical and phase coexistence data, and those for most soft materials (protein, polymers, surfactants) are implemented in several types of forcefields such as CHARMM 36\(^{63}\), AMBER\(^{64,65}\) and OPLS\(^{66}\). The parameters for cytochrome \( c \), decanoic acid, ethoxylated surfactants are taken from CHARMM\(^{63}\), which have been shown to provide simulation results that are comparable to experimental measurements\(^{67,68}\). Note, solid materials, such as silica, the torsion potentials are unnecessary due to the limited number of rotational degrees of freedom inherent in solid-state systems and thus, are not included in our simulations. As explained in section 1.2.4, the parameters for silica are taken from the silica model database developed by Heinz’s group\(^{50}\), which has been shown to accurately predict the aqueous interfacial properties of all types of silica\(^{42}\).

### 2.1.3 Periodic boundary conditions

In our MD simulation, periodic boundary conditions must be used for the calculation of Ewald nonbonded interaction. We adopt the minimum image convention\(^{69}\) as illustrated in Figure 2.4. The central simulation box is surrounded by the periodic replicas, all atoms are tracked via periodicity. For example, if an atom moves out of the boundary of the central simulation box, periodicity is used to determine this atom’s neighbors in the central cell (see figure 2-4).
Figure 2.4. Periodic boundary conditions. The central simulation box is surrounded by the replicas of itself. Atom $i$, $j$ and $k$ in the real simulation box are represented as a dark purple, dark orange and dark green sphere, respectively, and the replicas in surrounding images are shown as light-colored spheres. The dashed circle indicates the potential cutoff range for atom $i$. Each atom interacts with the nearest atom or image in the periodic array.

2.2 The Verlet Integration Algorithm

Positions and velocities of all atoms (configurations of all molecules) in the system are updated at each timestep by integrating the equation of motion (Newton’s law). The most widely used integration algorithm is based on Verlet algorithm\textsuperscript{70}, and it’s a progeny that produces computationally more efficient algorithms. One of the most popular algorithms are the \textit{leap-frog}\textsuperscript{71} and velocity Verlet (v-Verlet) methods\textsuperscript{72}. These two methods are both symplectic (designed for Hamiltonian based systems), time-reversible, and they are guaranteed to conserve the total energy of the system. The \textit{leap-frog} method uses positions and forces at time $t$, and velocities at time $t - \frac{1}{2} \Delta t$ to update positions and velocities for all atoms (see equations 2.9 and 2.10). Moreover, the v-Verlet method uses forces, positions and velocities at time $t$ as shown in equation 2.11 and 2.12.
The trajectories are of course similar between the two methods, however, the leap-frog method is computationally more efficient as fewer communication calls are required. For our simulations in fatty acid/silica system and surfactant/silica system, the leap-frog integrator implemented in GROMACS package\textsuperscript{73} will be adopted, as this method is both accurate and computationally efficient for self-assembly of a large number of short-chain amphiphiles. Although the v-Verlet method is more computationally demanding than the leap-frog method, because of its accuracy, it will be used to calculate single protein adsorption, where it is important to precisely determine the binding residues on protein.

\[
v\left(t + \frac{1}{2} \Delta t\right) = v\left(t - \frac{1}{2} \Delta t\right) + \frac{\Delta t}{m} F(t) \quad (2.9)
\]

\[
r\left(t + \frac{1}{2} \Delta t\right) = r(t) + v \Delta t \left(t + \frac{1}{2} \Delta t\right) \quad (2.10)
\]

\[
v\left(t + \frac{1}{2} \Delta t\right) = v(t) + \frac{\Delta t}{2m} F(t) \quad (2.11)
\]

\[
r(t + \Delta t) = r(t) + v \Delta t \left(t + \frac{1}{2} \Delta t\right) \quad (2.12)
\]

2.3 Simulations in an Ensemble

A thermodynamic state of a system is defined by macroscopic variables such as temperature \((T)\), pressure \((P)\), number of particles \((N)\) and total energy \((E)\).\textsuperscript{74} The microscopic state of a system is defined by atom positions and momenta in a phase space. A single point in the phase space described the one microscopic state of the system. An ensemble is a collection of these points in the phase space satisfying the conditions of a particular thermodynamic state. To make sure that all microscopic states generated in MD simulations are at the same thermodynamic state, the simulations must be performed in an ensemble. For our studies, we have a fixed number of

24
particles for each system, and we are interested in the microscopic properties at several fixed temperature points. We will focus on two ensembles: (i) a fixed number of particles, fixed volume, and fixed temperature \((NVT)\) ensemble and (ii) a fixed number of particles, fixed pressure, and fixed temperature \((NPT)\) ensemble. As shown in Figure 2.5a, the \(NVT\) ensemble represents the possible states of a mechanical system in thermal equilibrium with a thermal bath at a constant temperature. The system exchanges energy with the thermal bath, and the internal energy fluctuates to keep the temperature fixed. The \(NPT\) ensemble in Figure 2.5b is a mechanical ensemble that maintains constant temperature and pressure by coupling to a thermal bath and a “pressure bath”, where the volume and the internal energy fluctuate in order to keep both the pressure and temperature fixed. The thermal bath and “pressure bath” are referred to as the thermostat and barostat, respectively.

Figure 2.5. (a) An \(NVT\) ensemble connecting to a thermostat. Four possible states are presented to show that the number of particles, volume and temperature are fixed while that the internal energy of the system fluctuates. (b) An \(NPT\) ensemble coupling to the thermostat and barostat to maintain a fixed temperature and pressure. The exampled four states show that the internal energy and volume of the system fluctuate to stabilize the temperature and the pressure.
2.3.1 Thermostats and barostats

In MD simulations, the most widely used thermostat and barostat is the weak-coupling scheme of Berendsen\textsuperscript{75}. As shown in equation 2.13 and 2.14, the Berendsen coupling method mimics the first-order kinetics with a time constant $\tau$ to an external heat bath at a given temperature $T_0$, or an “pressure bath” at a given pressure $P_0$. While it can quickly attain the targeted temperature or pressure, it has been argued that it does not generate a real canonical ensemble as it suppresses the fluctuations of the kinetic energy. Thus, the Berendsen method can only be used at the beginning of the simulation to rescale the simulation box.

\[
\frac{dT}{dt} = \frac{T_0 - T}{\tau} \tag{2.13}
\]

\[
\frac{dP}{dt} = \frac{P_0 - P}{\tau} \tag{2.14}
\]

The velocity-rescaling (V-rescale) thermostat overcomes the shortcoming of Berendsen method and is adopt for our simulations for temperature coupling. The V-rescale method\textsuperscript{76} adds a stochastic term which ensures a correct kinetic energy distribution on the basis of Berendsen method, which is shown in equation 2.15, where $K$ is the kinetic energy, $N_f$ is the degrees of freedom. $dW$ is a Wiener process in the interval $[0, 1]$, which is a stochastic process consisting of random fluctuations of the kinetic energy.

\[
dK = (K_0 - K) \frac{dt}{\tau} + 2 \sqrt{\frac{K K_0}{N_f \sqrt{\tau}}} \tag{2.15}
\]

Parrinello-Rahman Barostat\textsuperscript{77} can be used for a more precise pressure coupling. In this method, the simulation box vector $\mathbf{b}$ obey the matrix equation of motion\textsuperscript{78}, as shown in equation 2.16,
\[ \frac{d\mathbf{b}^2}{dt^2} = VW^{-1}b^{t-1}(P - P_0) \] (2.16)

where \( V \) is the volume of the simulation box, \( W \) is the matrix parameter determining the strength of the pressure coupling, \( P \) stands for pressure, and \( P_0 \) is the reference pressure.

For our simulation, we use the v-rescale thermostat throughout to couple the temperature. For the pressure coupling, we first use weak-coupling method (Berendsen method) to rescale the volume of simulation box to get to the target pressure. Then we adopt the Parrinello-Rahman Barostat for the NPT simulation to obtain the dynamic equilibrium, which is determined via the time correlation function that is discussed in the next section.

2.3.2 Time averages and ensemble averages

The positions \( \mathbf{r} \) and momenta \( \mathbf{p} \) of all atoms can be represented in a phase space. For a system consisting of \( N \) atoms, \( \mathbf{r}^N \) and \( \mathbf{p}^N \) at any time defines a possible point/state in the \( 6N \)-dimensional phase space, and the system’s evolving state over time traces a trajectory. The time average of a property \( \mathcal{A}(\mathbf{r}^N, \mathbf{p}^N) \) over a period of time \( \tau \) can be defined as

\[
\langle \mathcal{A} \rangle_t = \lim_{\tau \to \infty} \frac{1}{\tau} \int_0^\tau [\mathcal{A}(\mathbf{r}^N, \mathbf{p}^N)] \, dt
\] (2.17)

In statistical mechanics, an ensemble is a probability distribution of the states of a system. The average property of a system can be obtained by calculating the ensemble average, which is the integration of the probability density of all states, \( f(\mathbf{r}^N, \mathbf{p}^N, t) \) at a fixed time, \( t \), as shown in equation 2.18.

\[
\langle \mathcal{A} \rangle_e = \int \int \mathcal{A}(\mathbf{r}^N, \mathbf{p}^N) \, f(\mathbf{r}^N, \mathbf{p}^N, t) \, d\mathbf{r}^N \, d\mathbf{p}^N
\] (2.18)

Sufficiently long-time integrations are typically necessary for the accurate simulation of a system in order to allow the system to go through all possible microstates \( (\mathbf{r}^N, \mathbf{p}^N) \). In fact, when
the probability density of all states \((f)\) is independent of time, and the ensemble average is equals to the time average in equation 2.19, the system can be considered to be equilibrated.

\[
\langle \mathcal{A} \rangle_e = \langle \mathcal{A} \rangle_t
\]

(2.19)

This equivalence is known as the ergodic theorem. To determine if a simulation is ergodic, several macroscopic average properties (i.e. temperature, total energy, density) as well as the microstates (position of all atoms) can be evaluated. The time-independence of the microstates can be checked by calculating time correlation function, which is defined as root mean square deviation (RMSD) between a configuration at time \(t\) with respect to their initial configurations (see equation 2.20)

\[
RMSD(t) = \left[ \frac{1}{M} \sum_{i=1}^{N} m_i ||r_i(t) - r_i(t_0)||^2 \right]^{1/2}
\]

(2.20)

where \(m_i\) is the mass of atom \(i\), \(M = \sum_{i=1}^{N} m_i\), \(r_i(t_0)\) is the position of atom \(i\) in initial configuration at \(t = t_0\) and \(r_i(t)\) is the position of atom \(i\) at a later time \(t\). The RMSD will converge to a constant if the configurations of all atoms are no longer correlated with \(t\). Therefore, the equilibrium state of our system is reached when temperature, density and RMSD is independent of time.
CHAPTER THREE
REORIENTATION OF CYTOCHROME C ON IONIZED SILICA SURFACES†

3.1 Introduction

Protein-inorganic surface interactions involved in the protein-solid surface system are gaining more and more attention due to their occurrence in a broad range of applications in nanotechnology, such as biosensors\textsuperscript{14}, bioreactors, and bioanalytical devices. As addressed in section 1.2.1, the protein is expected to be constrained on the surface with preferred orientations to accommodate various needs, and the performances of these devices will be highly influenced if the protein undergoes significant spatial reorientation or conformational deviation from the preferred configuration. Thus, it is essential to develop an understanding of the factors that affect the protein-inorganic surface interactions that direct the adsorption and reorientation process. The interactions of protein molecules with inorganic materials (silica in this study) are determined not only by the protein molecules but also by the silica surface's physicochemical properties in water media, such as the chemical states and the shape of the silica materials. In this chapter, we will show that the degree of the ionization of the silica surface, $I_{\text{silica}}$ influences the electrostatic interaction between a globular protein, cytochrome $c$ and the silica surface, governing the final adsorbed state of the protein molecule.

Cytochrome $c$ (cyt $c$) is a small globular protein of 104 amino acid residues covalently bound to a central heme group, with five long $\alpha$-helixes (denoted as $\alpha_A$-$\alpha_E$), a short $3_{10}$ helix, and two parallel $\beta$-sheets connected by loops as shown in Figure 3.1a\textsuperscript{79,80}. It plays a central role in the oxygen transport and ATP synthesis in mammals. It is widely considered as a “hard” protein that

\textsuperscript{†} Section 3.2, Figure 3.1a, Figure 3.3 and Table 3.1 have been published on the Soft Matter journal, and they are reproduced from Ref.\textsuperscript{67} with permission from The Royal Society of Chemistry.
retains its ellipsoid-like shape \((2.6 \times 3.0 \times 3.4 \text{ nm})\). As shown in Figure 3.1c, the protein surface is colored by residue charges. There are 21 positively charged residues (19 Lysine plus 2 Arginine residues) and 12 negatively charged residues (3 Aspartic plus 9 Glutamic acids). Thus, cyt \(c\) carries the net charge of \(+9e\) at pH 7 with an isoelectric point at pH 10. However, the charged amino acid groups are not uniformly distributed over the protein surface. To predict the adsorption sites and the resulting orientation of cyt \(c\) on silica surface that has both SiOH and SiO⋯Na\(^+\) groups, it is unsound by solely locating the positively charged residues, hydrophilicity (polar groups) of the protein should also be considered. The protein is hydrophilic (blue region in Figure 3.1b) at the top right and bottom left region, and in the central part, both hydrophilic and hydrophobic

![Figure 3.1. Structure of protein cytochrome \(c\) with different coloring method. (a) Protein surface colored by residue charge is shown as a ghost surface. The secondary structure is displayed by solid ribbon, where five major \(\alpha\)-helices are colored in purple, the 310 extending helix is in colored lime, pi-helix is in red, extended \(\beta\)-sheets are in blue, \(\beta\)-turns are in pink and the bridged \(\beta\)-sheet is in cyan. N-terminus and C-terminus is indicated by red and purple licorice, respectively, and heme ligand is shown in pink. Lys, Arg and Gln residues are colored by deep blue, yellow and black, respectively. (b) The protein surface is colored by hydrophobicity of residues, where hydrophobic (nonpolar) and hydrophilic (polar) groups are colored in red and blue, respectively. (c) The protein surface is colored by the charge of residues, where positive residues are colored in blue, negative ones are shown in red and neutral ones are in white.](image-url)
groups are observed with hydrophobic region predominant. By conducting all-atom molecular dynamics, we found that the electrostatic interaction between the cyt c and silica increases as a function of the degree of ionization of the silica surface, driving the cyt c to be adsorbed via more residues and reorientating the protein from a “head-on” to a “side-on” configuration. Further, the adsorbed cyt c remains in its native folded conformation, suggesting its structural stability after adsorption.

3.2 Simulation Setup and Methods

To explore the orientation of cyt c on silica nanoparticles as a function of the degree of silica’s ionization, we conducted four simulations to simulate the adsorption process of a cyt c on silica surfaces of four degrees of ionization ($I_{\text{silica}}$), namely 5 %, 9 %, 18 %, and 27 %, and the density of silanol groups of the silica surface is 4.7 nm$^{-2}$. These $I_{\text{silica}}$ values are chosen to model the silica/water interface near pH 3, 5, 7 and 9. (See section 1.2.4) The upper limit is at pH of 9 above which the silica will be dissolved. The simulation box setup is shown in Figure 3.2.

The silica surface model was constructed by following Heinz’s study$^{50}$. As explained in section 1.2.4, the surface of silica particles of medium size is mimicked by using the Q$_3$/Q$_4$ silica surface model with a total density of SiO(H, Na$^+$) groups of 4.7 nm$^{-2}$. This model was obtained by the cleavage of the (10\overline{1}) plane of $\alpha$-cristobalite and the hydration of dissociated bonds to silanol groups. Silica surfaces with various $I_{\text{silica}}$ values are constructed by the deletion of hydrogen atoms from $\equiv$Si-OH groups (silanol groups) and the addition of sodium ions to create $\equiv$SiO$^- \cdots $Na$^+$ groups (siloxide groups). This model has been shown to accurately predict the aqueous interfacial properties by extensive comparisons to experimental measurements$^{42}$.

2giw.pdb$^{82}$ from RCSB Protein Data Bank was used as the starting cyt c structure. First, a solvated cyt c was prepared to resemble the cellular environment more closely. The solvation box,
Figure 3.2. Illustration of the simulation box adopting the periodic boundary condition (PBC). The surface at the degree of ionization of 27% is used as a representative to show the topology of the silica slab, where the silicon is shown in yellow, oxygen is in red, hydrogen is in white and sodium ion is in pink. The protein solvated in the center of a TIP3P water box is placed above the silica slab for adsorption study. The boundary of the simulation box is demonstrated by the black rectangle. The protein can also be adsorbed to the upper or bottom surface of the silica slab due to the periodic boundary condition. The coloring method of the cyt c follows the way in Figure 3.1a. referred to as the protein/water space in Figure 3.2, is used as the initial configuration for all four simulations. The protein is placed in the center of a water box of dimension $100 \, \text{Å} \times 104.5 \, \text{Å} \times 85 \, \text{Å}$ using TIP3P as the water model, and then it was neutralized by sodium chloride (NaCl) at an ionic strength of 0.02 M. Energy minimization was performed to remove the potential overlap of water molecules, and ions by using the steepest descent algorithm. The parameters for cyt c were adopted from the CHARMM36 forcefield database for protein. This solvation box is then placed above the silica surface at different degrees of ionization, respectively, to construct the
simulation box for adsorption study, as shown in Figure 3.2. The simulation box is represented by a black rectangular frame. The protein can also be adsorbed to the upper or bottom surface of the silica slab due to the applied periodic boundary condition (PBC). (see section 2.1.3)

After the setup of the simulation box, energy minimization was again performed for the whole system comprising silica surface, protein, water, sodium, and chloride ions to avoid intermolecular clashes between silica surface and water. The simulation box was heated up to 300 K at the rate of 0.1 ps/K, followed by the equilibration for 270 ps to stabilize the temperature to 300 K and the pressure to 1 bar. (see section 2.3) The equilibration simulation was performed for a 60-100 ns long run at 300 K in an NPT ensemble using periodic boundary conditions until the protein was adsorbed and stabilized on silica surface (see section 2.1.3). Silicon and oxygen of silica slab were fixed throughout the simulation by following the common recipe for the protein adsorption studies\(^{86,87}\), only protein, water, and ions were flexible. The integration timestep was 1 fs. The cutoff for van der Waals interactions was 12 Å, and the smooth particle mesh Ewald (SPME) summation with PME grid spacing ratio of 1 was used for the calculation of the electrostatic interaction.

### 3.3 Orientation of the Adsorbed Cytochrome C

In all four simulation runs, the cyt c was adsorbed to the silica surface within 100 ns. The protein is considered to be adsorbed when the distance between silica and protein is below 5 Å.\(^{86}\) The snapshots of the adsorbed protein on silica surface as a function of the degree of ionization, \(I_{\text{silica}}\) is shown in Figure 3.3. Given that cyt c can be considered as an ellipsoid-like “hard” protein, the difference in the relative position of adsorbed protein to the silica surface can be directly characterized by the change in its height, \(D_n\). \(D_n\) is measured and averaged over the equilibrium
Figure 3.3. Equilibrium adsorption state of cytochrome c on silica surface of an increasing degree of ionization, $I_{\text{silica}}$. SiO$_2$ are shown by space-filling method where silicon is yellow, oxygen is red, hydrogen is white, and ions are pink. Water molecules are not shown for clarity. The representation method of protein is the same as that in Figure 3.1a.

trajectory and plotted in Figure 3.4b, and the maximum height ($D_{n,max}$) is denoted as a dashed reference line in Figure 3.3. The decrease of the height of the adsorbed protein at an increasing $I_{\text{silica}}$ indicates its orientational change induced by the external silica surface.

To characterize the exact orientation of the adsorbed ellipsoid-like cyt c, the angle between its long axis and the surface normal, $\theta_n$, is tracked throughout the equilibrium trajectory. The long axis is defined as the line connecting the C$_\alpha$ atom of Gly1 (C-terminus) and N atom of Lys 53 ($\alpha_B$ helix). As illustrated in Figure 3.4a, the orientation of the adsorbed protein can be classified into three types, namely, (i) “head-on” orientation ($0 \leq \theta_n < 30^\circ$), (ii) “side-on” orientation ($60^\circ \leq \theta_n \leq 90^\circ$) and (iii) the orientation “between” these two ($30^\circ \leq \theta_n < 60^\circ$). The orientation distribution of adsorbed protein on different silica surfaces is shown in Figure 3.4d, and the three regions divided by the dashed lines denote those three possible orientations. The mean orientation angle, $\theta_{n,av}$, is calculated and plotted as a function of $I_{\text{silica}}$ in Figure 3.4c.

The average orientation angle in Figure 3.4c increases from $24^\circ$ to $70^\circ$ as the degree of
Figure 3.4. (a) Schematics illustrating three orientation types of an adsorbed protein, namely (i) “head-on” orientation, (ii) “between” orientation and (iii) “side-on” orientation. The orientation is defined as the angle between long axis of protein and silica surface normal, $\theta_n$. (b) Average height of adsorbed protein referenced to silica surface, $D_n$, and (c) Average orientation of adsorbed protein, $\theta_{n,av}$ as function of the degree of ionization of silica. (d) Distribution of the orientation angle of the adsorbed protein over the last 10-ns equilibrium trajectory. The orientation angle is normalized in the way that the integration over the curve is 1. The protein is adsorbed as a “head-on” orientation at a weakly charged silica surface, and it transforms to a “side-on” orientation as the degree of ionization is increased. The distribution of the orientation shows that the position of the adsorbed protein fluctuates in a narrow range, implying the stability of a certain orientation of the adsorbed protein on a certain surface. The stability of the adsorbed protein is higher on the silica surface of a higher degree of ionization.

ionization of silica surface ($I_{\text{silica}}$) is increased from 5 % to 9 %, indicating that the protein reorientates from a “head-on” to a “side-on” configuration (Figure 3.4a). In Figure 3.4d, a similar bimodal distribution is observed for the adsorbed protein at $I_{\text{silica}}$ of 5 % and 9 %, which has a major peak followed by a minor peak. For $I_{\text{silica}}$ of 5 %, the protein spends most of the time to be adsorbed in a “head-on” orientation. Besides, it has a probability of 16 % (integration from 30° to
60°) to be adsorbed in a “between” orientation, indicating that the protein oscillates between these two states. On the other hand, at $I_{\text{silica}} = 9\%$, both major and minor peaks occur in the “side-on” region, and the broad peaks demonstrate the instability of the adsorbed protein. Further, as $I_{\text{silica}}$ is increased from 9 % to 18% and 27 %, only one major peak occurs in the “side-on” region, and the peak is getting narrower. The narrower major peak implies that the protein is more stably adsorbed in the “side-on” orientation as the silica surface is more ionized. The origin of the orientation transition and the stability of the characteristic orientation at different $I_{\text{silica}}$ should be related to the binding sites and the electrostatic interaction between the protein and silica surface, which is discussed in the followed section.

3.4 Binding Mechanism of the Cytochrome C

To fully understand the binding mechanism of cyt c on the silica surface carrying different numbers of negative charges, we first find out the residues that act as binding anchors. The residue is considered as a binding anchor if the distance between the residue and the surface is smaller than 5 Å. The binding anchors are summarized in Table 3.1. We found that the binding face on the protein is located in the hydrophilic region (blue region in Figure 3.1b). The most important anchor residues contributing to the adsorption of cyt c are positively charged residues, Lysine (Lys), and polar neutral residues, Glutamine (Gln). The binding of the protein occurs via protonated amino groups ($\text{NH}_3^+$) in Lysine and polar amino groups ($\text{NH}_2^-$) in Glutamine to negatively charged siloxide groups ($\equiv \text{SiO}^{-} \text{Na}^+$).

As the total surface area of silica slab is fixed, the increase of the siloxide groups’ density directly reflects the increase of the absolute number of the negatively charged siloxide groups. In Table 3.1, it can be found that at $I_{\text{silica}}$ of 5 %, the silica surface is weakly ionized, and the discrete
Table 3.1. Binding anchors of cyt c adsorbed on silica surfaces of different degrees of ionization.

<table>
<thead>
<tr>
<th>Degree of ionization of silica, $I_{\text{silica}}$</th>
<th>Density of siloxide groups ($\equiv \text{SiO}^-\text{Na}^+$) ($\text{nm}^{-2}$)</th>
<th>Preferred orientation</th>
<th>Anchored residues</th>
</tr>
</thead>
<tbody>
<tr>
<td>5 %</td>
<td>0.235</td>
<td>Head-on</td>
<td>Lys8, Lys5</td>
</tr>
<tr>
<td>9 %</td>
<td>0.423</td>
<td>Side-on</td>
<td>Lys79, Lys27, Lys25</td>
</tr>
<tr>
<td>18 %</td>
<td>0.846</td>
<td>Side-on</td>
<td>Lys87, Lys86, Gln16, Lys79, Lys8</td>
</tr>
<tr>
<td>27 %</td>
<td>1.269</td>
<td>Side-on</td>
<td>Gln12, Lys86, Lys87, Lys8, Lys5</td>
</tr>
</tbody>
</table>

Charges at the surface dominate the binding of the protein. The sparsely distributed charged sites on silica drive the ellipsoidal protein’s binding with only two amino acid residues (Lys8 and Lys5) (Figure 3.3a). The limited number of binding sites also accounts for the relative instability of cyt c, leading to its oscillation from “head-on” to “between” orientation states (Figure 3.4d). As $I_{\text{silica}}$ is increased to 9%, the number of siloxide groups ($\equiv \text{SiO}^-\text{Na}^+$) increases, and the protein binding occurs via three major charged sites, namely Lys79, Lys27, and Lys25. The increasing number of binding sites on silica drives the adsorption of protein via multiply charged amino acid residues and thus reorients cyt c from a “head-on” to a “side-on” state. As the silica surface is further ionized, the negative charge density is increased, enabling the hydrogen atom in polar groups ($\text{NH}_2^+$) to be bound to the surface. The increasing number of binding sites at $I_{\text{silica}}$ of 18% and 27% not only immobilize the protein in a “side-on” orientation, but also increase the stability of such adsorbed state (see the narrow peak in Figure 3.4c). The correlation between the number of charged binding sites and the different adsorbed states suggests that the driving force for the adsorption is
electrostatic interaction. Herein, the charge-charge electrostatic interaction between the cyt c and the silica, \( E_{elec} \), is extracted and plotted as a function of \( I_{silica} \) in Figure 3.5. \( E_{elec} \) increases with the increasing negative charges on the silica surface, leading to increased binding sites on the protein. Hence, the enhanced electrostatic attraction initiates the protein to reorientate from “head-on” to “side-on” adsorbed state.

![Graph showing electrostatic interaction energy between silica and cyt c](image)

Figure 3.5 Electrostatic interaction energy, \( E_{elec} \), between adsorbed cyt c and the silica surface of different degrees of ionization, \( I_{silica} \). \( E_{elec} \) increases monotonically with \( I_{silica} \), leading to increased binding residues on cyt c.

### 3.5 Structural Stability of the Adsorbed Cytochrome C

The protein’s geometric shape, also known as its tertiary structure, is determined by its backbone (NH–C\( \alpha \)H–C=O).\(^{88}\) In order to examine the structural stability of the adsorbed protein on silica surfaces of different degrees of ionization, the radius of gyration of the backbone, \( R_{g,b} \), is extracted from the last 5-ns of the equilibrium trajectory, as shown in Figure 3.6. The radius of
gyration is a common property used to characterize the compactness and folded/unfolded states of the protein structure, and it can be calculated via equation \(3.1\):

\[
R_{g,b} = \left( \frac{\sum_i \| \mathbf{r}_{b,i} \|^2 m_{b,i}}{\sum_i m_{b,i}} \right)^{1/2}
\]

where \(m_i\) is the mass of atom \(i\) in protein’s backbone, and \(\mathbf{r}_{b,i}\) is the relative distance of atom \(i\) with respect to the center of mass (COM) of the protein.

Figure 3.6. Radius of gyration, \(R_{g,b}\) of the backbone of adsorbed cyt \(c\) on silica surface of different degrees of ionization over the equilibrium trajectory. The horizontal dashed lines indicate different tertiary structures of cyt \(c\), where the orange one represents the folded state and the black one stands for the unfolded state. \(R_{g,b}\) of cyt \(c\) on all silica surfaces fluctuate near 13 Å, and the maximum variance is within 0.5 Å, implying the structural stability of cyt \(c\) after adsorption.

The \(R_{g,b}\) value for the original cyt \(c\) in a fully folded state is 12.54 Å, and that in an unfolded state is 18.94 Å. The folded and unfolded state of cyt \(c\) is marked as the orange and black dashed line in Figure 3.6, respectively. We found that the \(R_{g,b}\) values of adsorbed cyt \(c\) on all silica surfaces studied here is relatively stable with minor fluctuation near its folded state.
(orange dashed line), also referred to as its native conformation\(^\text{13}\). In addition, the \( R_{g,b} \) value for the adsorbed cyt at each \( I_{\text{silica}} \) shows a variation within 0.5 Å, implying that cyt c maintains its original geometry shape after adsorption on all silica surfaces. Thus, cyt c can be adsorbed on the silica surface with different orientations while maintaining its native conformation.

### 3.6 Summary

This chapter shows that the charge-charge electrostatic interaction is responsible for the reorientation of cytochrome c. The electrostatic interaction is shown to be modulated via the degree of ionization of the silica surface, \( I_{\text{silica}} \), with a positive correlation, leading the cyt c to be adsorbed on the silica surface with a higher number of binding residues. The increased number of anchored residues on cyt c leads the protein to reorientate from a “head-on” to a “side-on” adsorbed state with its native conformation preserved. This study can be further validated by similar studies on the protein molecules’ preferential binding, both experimentally and computationally. For instance, studies have shown that human carbonic anhydrase and lysozyme are bound to the silica surface via positive residues driven by electrostatic interactions.\(^{91,92}\) Most importantly, we show that the protein-adsorbent electrostatic interaction can be programmed by the external parameter, \( I_{\text{silica}} \), to modulate the adsorbed states. In addition to the degree of ionization of the adsorbent, the protein-adsorbent electrostatic interaction can also be changed by modifying the adsorbents with different terminal functional groups such as –COOH, –NH\(_2\), –SO\(_3\)H, and –OH.\(^{93}\) This study presents a basis for understanding the role of protein-adsorbent electrostatic interaction in driving the discrete protein molecules in different orientations, enabling a design-principle of nanomaterial fabrication for biomedical applications such as biosensors and bioanalytical devices.
CHAPTER FOUR
SELF-ASSEMBLY OF FATTY ACIDS ON AMINE-FUNCTIONALIZED SILICA SURFACES†

4.1 Introduction

Fatty acids are natural carboxylic acids that belong to the antimicrobial agents, and they have wide applications in detergency, cosmetics, and biological techniques. The study of the morphology and phase behavior of fatty acids is essential in utilizing their functionalities to develop and optimize these applications. For example, it has been reported that the formation of bilayers/vesicles by fatty acids can be considered as a possible step towards the hypothetical protocells.94–96 Fatty acid-based bilayers/vesicles can also be applied for enzyme-catalyzed reactions.28 However, the development of those applications still has concerns regarding the structural stability, as fatty acids in aqueous solution are overly sensitive to pH, leading to difficulty in guiding the system to the desired structure. Thus, it is significant to understand the effect of pH on the molecular interactions that decide the morphology and phase behavior of the fatty acid assemblies.

While the self-assembly process of fatty acid in aqueous bulk solution has been widely studied, the adsorbed states and phase behavior of fatty acid assemblies at the solid-water interface are not fully understood. In aqueous bulk solutions, the self-assembly state and phase behavior of fatty acids are decided by the van der Waals interaction between alkyl chains and the electrostatic interaction between the polar groups at different ionization states under certain acid/basic environment (pH). The balance between these two interactions determines that fatty acid

‡Table 4.1, Figure 4.3b, Figure 4.4 and Figure 4.6a previously published in Ref.111 are reproduced here with the permission from the American Chemical Society (ACS). The article can be found through https://pubs.acs.org/doi/abs/10.1021/acslangmuir.0c00156, and further permissions related to the material excerpted should be directed to the ACS.
molecules form micelles at high pH values, bilayer/vesicles at intermediate pH values, and oil phase at low pH values in the aqueous solution. (see section 1.2.2) By introducing the propyl ammonium functionalized silica surface ($m$SiO$_2$), the molecular mechanism responsible for the reconfiguration of fatty assembly and its corresponding phase behavior is proposed to be the competition between fatty acid-surface interaction and fatty acid-fatty acid interaction. In this study, we adopt molecular dynamics to understand the response of these two interactions as a function of the pH of decanoic acid (C10) solution. That determines the morphology and phase behavior of adsorbed decanoic acid assemblies at $m$SiO$_2$-water interfaces.

By definition$^{97}$, pH is a scale used to specify the molar ratio of the free hydrogen ions, which is referred to as the degree of deprotonation of fatty acid molecules, $R$. We found that as a function of $R$, the electrostatic interaction between $m$SiO$_2$ and decanoic acids is enhanced while van der Waals interaction among decanoic acid molecules are weakened. The competition between these two interactions leads to different morphologies of adsorbed decanoic acid assemblies at different $R$ values. In the region of $R < 0.3$ (low pH), the electrostatic attraction from $m$SiO$_2$ is weak, the decanoic acid molecules form oil droplets at $m$SiO$_2$-water interfaces, resembling their phase behavior in bulk solution. On the other hand, as $R$ is increased well above 0.5 (high pH), the electrostatic attraction from $m$SiO$_2$ starts to dominate, and the bilayer structure is observed. The formation of bilayer at high $R$ is different from the phase behavior in the aqueous bulk solution where globular micelles are formed. Based on this mechanism, experimentalists can control the formation of bilayer structure by adjusting the degree of deprotonation of the fatty acid solution to a wider range ($R > 0.5$), compared to the previous narrow range of $R \approx 0.5$ in dilute alkaline solutions.
4.2 Simulation Model and Setups

To model the spontaneous self-assembly of decanoic acid molecules on a positively charged surface at room temperature, an alkylammonium-modified silica surface ($mSiO_2$) was constructed. As discussed in section 1.2.4, the use of alkylammonium functional group on the silica surface is widely used to provide a compatible charged surface for biomaterial adsorption. First, a silica surface was obtained by the cleavage of (001) plane of $\alpha$-quartz and the hydration of dissociated bonds to silanol groups. Next, a selection of 180 homogenously dispersed hydroxyl groups were replaced by the protonated alkylamine functional groups in a monodentate manner, leading to the formation of Si-C bonds as shown in Figure 4.1c.

![Figure 4.1](image)

Figure 4.1. Atomic model of (a) decanoic acid molecule and (b) propyl ammonium functionalized silica surface ($mSiO_2$). (c) Zoomed-in perspective view of $mSiO_2$ showing the configuration of propyl ammonium functional groups. Decanoic acid monomer and propyl ammonium functional group are displayed in ball-and-stick style. Carbon, oxygen, hydrogen, and silicon atoms are colored in yellow, red and white bonds, respectively.

After the functionalization of silica surface, we need to establish a stable $mSiO_2$-water interface by performing energy minimization for both $mSiO_2$ structure and $mSiO_2$/water system. Parameters in both bonded and nonbonded potential functions (see Chapter 2) for the bare silica
are adopted from previous studies\textsuperscript{50}, and that for propylammonium groups are from CHARMM general forcefield\textsuperscript{98}. Energy minimization was performed to relax the structure of $m$SiO$_2$ at $T = 0$. Next, the aqueous solution containing chloride ions was added to the $m$SiO$_2$ for neutralization, and energy minimization for $m$SiO$_2$/water system was performed using steepest descent algorithm\textsuperscript{83} with no temperature assigned until the potential of the simulation box converged to a constant.

After obtaining an optimized structure of $m$SiO$_2$ surface, we constructed the decanoic acid/$m$SiO$_2$/water system to model the self-assembly and adsorption decanoic acid to $m$SiO$_2$ surface. The configuration of decanoic acid (C10) molecules is displayed in Figure 4.1a, where the C$_1$ and C$_{10}$ atoms in the alkyl chain are marked. A fixed number of decanoic acid molecules (100) with a various molar ratio of deprotonated molecules, defined as $R = \frac{N_{\text{COO}^-}}{N_{\text{COOH}} + N_{\text{COO}^-}}$, also known as the degree of deprotonation, were placed above the $m$SiO$_2$ surface. Protonated ethanolamine was used as the counterion to neutralize the deprotonated decanoic acid. Parameters in both bonded and nonbonded potential functions for the decanoic acid are adopted from CHARMM-lipid forcefield\textsuperscript{99}, and that for ethanolamine molecules are from CHARMM general forcefield\textsuperscript{98}. The details on the solution composition are listed in Table 4.1, and $R$ was systematically increased from 0.1 to 1, featuring the pH from below to above the $pK_a$ as the pH condition of the solution is proportional to the $R$ value. The concentration of all fatty acid molecules (both protonated and deprotonated) in all systems is 110 $mM$. In a typical simulation box, a mixture of protonated decanoic acid and deprotonated decanoic acid with protonated ethanolamine molecules corresponding to different $R$ values were placed above the $m$SiO$_2$ surface. The initial configuration of the mixture was created by Packmol package\textsuperscript{100}, which is a powerful tool to minimize the possible overlap of atoms’ coordinates. The mixture together with the $m$SiO$_2$ was then solvated in a water box of size 100×104×130 Å$^3$ using the TIP3P model\textsuperscript{101}. As a reference, the self-assembly of decanoic acid
molecules in bulk water (without $m\text{SiO}_2$) is also simulated at 298 $K$ as a function of $R$, the composition is the same as displayed in Table 4.1.

Table 4.1. The number of deprotonated decanoic acid molecules ($N_{\text{COO}^-}$), protonated decanoic acid molecules ($N_{\text{COOH}}$), counterion ($N_{\text{HOCH}_2\text{CH}_2\text{NH}_3^+}$) and water molecules ($N_{\text{H}_2\text{O}}$) in the simulation box. The composition of fatty acid solution indicates the pH environment of fatty acid solutions.

<table>
<thead>
<tr>
<th>pH environment</th>
<th>Degree of deprotonation of fatty acid, $R = \frac{N_{\text{COO}^-}}{N_{\text{COOH}} + N_{\text{COO}^-}}$</th>
<th>$N_{\text{COO}^-}$</th>
<th>$N_{\text{COOH}}$</th>
<th>$N_{\text{HOCH}_2\text{CH}_2\text{NH}_3^+}$</th>
<th>$N_{\text{H}_2\text{O}}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH &lt; $pK_a$</td>
<td>0.1</td>
<td>10</td>
<td>90</td>
<td>10</td>
<td>48560</td>
</tr>
<tr>
<td>pH ≈ $pK_a$</td>
<td>0.2</td>
<td>20</td>
<td>80</td>
<td>20</td>
<td>48520</td>
</tr>
<tr>
<td>pH &gt; $pK_a$</td>
<td>0.3</td>
<td>30</td>
<td>70</td>
<td>30</td>
<td>48480</td>
</tr>
<tr>
<td>pH ≈ $pK_a$</td>
<td>0.5</td>
<td>50</td>
<td>50</td>
<td>50</td>
<td>48400</td>
</tr>
<tr>
<td>pH &gt; $pK_a$</td>
<td>0.7</td>
<td>70</td>
<td>30</td>
<td>70</td>
<td>48000</td>
</tr>
</tbody>
</table>

Energy minimization was performed to remove bad intermolecular contacts (atomic overlap) using steepest descent algorithm$^{83}$, and a good initial configuration of the solution can be obtained when the potential energy converges to a constant. The NVT pre-equilibration using V-rescale thermostat$^{102}$ was performed to stabilize the temperature of simulation box to 298 K. Next, to stabilize the pressure to 1 bar, simulations were carried out in an NPT ensemble, where the Berendsen barostat$^{75}$ was applied to quickly rescale the size of the simulation box at 1 bar. Finally, the long equilibration simulations were performed in the NPT ensemble using V-rescale thermostat
and Parrinello-Rahman barostat\textsuperscript{103} for at least 20 ns. Particle-Particle Particle-Mesh algorithm (P3M-AD)\textsuperscript{104} based on Ewald summation (see section 2.1.1) is used for long-range electrostatic interactions, and that is for charge-charge interactions between nonbonded atoms with partial charges. The leap-frog algorithm\textsuperscript{71} with a time step of 0.1 fs was used to integrate the equation of motion, and the cutoff for vDW interactions and the real-space of Ewald sum was set to 1.2 nm. (see section 2.2) All analysis is based on the last 1 ns of the generated equilibration trajectory file. It is noted that for the evaluation of the shape and orientation of the fatty acid molecules throughout this chapter, only conditions at $R = 0.1, 0.3, 0.5$ and 1.0 are given to characterize the pH condition from below to above $pK_a$, and the plots for all $R$ values are provided in Appendix A.

**4.3 Phase Behavior of Decanoic Acid in Bulk Water at 298 K**

As a reference, the equilibrated assembled states of fatty acid molecules in bulk water at 298 K is displayed in Figure 4.2a-f. Only fatty acid aggregates are shown, the removal of the unassembled monomers, water and ions is for better visualization. It is observed that the fatty acid molecules form bulky aggregates with protonated headgroups exposed at low $R$ values ($R < 0.5$), bilayer at $R = 0.5$ and spherical micelles at high $R$ values ($R > 0.5$). The alkyl chain length is an indicator of the hydrated state of the fatty acid monomer in water. Thus, we examine the alkyl chain length distribution, defined as the distance between $C_1$ and $C_{10}$ atoms, as shown in Figure 4.2g and Figure A1. At all $R$ values, the distribution is trimodal, where the third sharp mode at 11.4 Å features a fully extended alkyl chain, and the shortening of the first two modes (near 9.8 Å and 10.5 Å) is due the tilting of $C_1$ and $C_{10}$ atoms. The intensity of the first two modes (shorter length) are much higher than the third mode at 11.3 Å, indicating that most fatty acid molecules are in contracted configurations, and the assemblies are in fluid phase. As $R$ is increased from
below to above 0.5, the density of alkyl chain length near 10.5 Å and 11.3 Å decrease remarkably, while the peak near 9.8 Å broadens and further shifts to a lower value. This suggests that the fatty

Figure 4.2. (a-f) Snapshots of equilibrated assembled state of fatty acid molecules in bulk water at room temperature as a function of deprotonated degree, $R$, increasing from 0.1 to 1.0. For decanoic acid molecules, hydrogen atoms are hidden. The alkyl chains drawn as blue licorice bonds, the oxygen in the deprotonated molecule is shown as the red sphere, and that in the protonated molecule is in pink. Water and counterions are not shown for clarity. Fatty acid molecules undergo bilayer-to-micelle transformation as $R$ increasing from 0.5 to 1.0, which agrees well with previous studies\textsuperscript{23,27,28,105–107}. (g) Probability distribution of the alkyl chain length (defined as C\textsubscript{1}-C\textsubscript{10} distance). The cumulative probability (integration of the probability density) of all curves is the same.
acid molecules further bend as the deprotonated degree is increased, and the deprotonated molecules in micelles (panel f) are in bent configurations. Together with the morphologies in the snapshots, it is concluded that in bulk water at room temperature, decanoic acid molecules (C10) of concentration 110 mM form oil droplets at low R values, fluid bilayers near $R = 0.5$ and micelles at $R > 0.5$. This observation agrees well with previous studies on the phase behavior of decanoic acids28,105–107 (C10), lauric acids27,29 (C12) and oleic acid23,24 (C18) near 100 mM at room temperature both computationally and experimentally. (also see section 1.2.2)

4.4 Morphology and Phase Behavior of Decanoic Acids at $m$SiO$_2$-Water Interfaces

For our study, the fatty acid-$m$SiO$_2$ electrostatic interaction and fatty acid-fatty acid van der Waals interactions are studied as a function of $R$ at room temperature to determine its effect on the resulted morphology and phase behavior of fatty acid assemblies at the $m$SiO$_2$-water interfaces. Both adsorbed assemblies, unadsorbed assemblies and unadsorbed monomers are observed, however, for this study we only focus on the adsorbed assemblies.

The aggregation number, $N_i$, and the carried negative charges, $Q_i$ for each assembly as a function of $R$ are extracted and mapped in Figure 4.3a. Each scatter in Figure 4.3a represents one assembly. The aggregation number ($N_i$) is defined as the number of fatty acid monomers comprising one assembly. The negative symbol of $Q_i$ means that the assemblies carry negative charges, and the value is equal to the number of deprotonated fatty acid molecules in assembly $i$. The mean height of all adsorbed assemblies referenced to the bare silica surface is also calculated and plotted as a function of $R$ in Figure 4.3b, which coarsely characterizes the size and growth pattern of all adsorbed assemblies at different degrees of deprotonations.

As illustrated in the distribution map in Figure 4.3a, in low $R$ region ($R < 0.4$), the adsorbed assemblies carry fewer negative charges (in magnitude) and are highly-aggregated, while
in the high $R$ region ($R \geq 0.7$), the adsorbed assemblies carry more negative charges (in magnitude) and exist as smaller aggregates. Thus, the increased number of deprotonated fatty acid molecules are distributed to several smaller assemblies due to the wide distribution of positive charges on the $mSiO_2$ surface. Additionally, the mean height of the assemblies decreases as a function of $R$, demonstrating that the assemblies spread out over the $mSiO_2$ surface at higher $R$ values.

Figure 4.3. (a) 2D aggregation number-assembly charge map extracted from the equilibrated states of fatty acid assemblies adsorbed on $mSiO_2$. Each scatter represents the adsorbed assembly $i$ showing its aggregation number, $N_i$ and the negative charges it carries, $Q_i$. (b) Mean height of adsorbed fatty acid assemblies referenced to bare silica, $D_h$ as a function of $R$, and the inset is the schematic representation of $D_h$.

In Figure 4.4, we display the equilibrated state of adsorbed fatty acid assemblies at each $R$ value, which describes the shape and the phase behavior of the adsorbed assemblies. Fatty acids can be adsorbed on both top and bottom sides of the $mSiO_2$ surface, and the side with majority assemblies adsorbed is shown. Water and ions are not shown for clarity. From Figure 4.3a we acknowledge that the deprotonated fatty acid molecules are not evenly distributed to all the assemblies in each simulation box, i.e. the local $R$ value for each assembly can be different from the global $R$ value of the solution. Thus, to accurately characterize the effect of $R$ on the
configuration of the adsorbed assembly, we define the assembly whose deprotonated degree is equal to the deprotonated degree of the whole system at each \( R \) as the characteristic assembly, as pointed by the arrow in Figure 4.4. For example, the characteristic assembly in simulation box of \( R = 0.1 \) has 10\% of the monomers deprotonated.

Figure 4.4. (a-f) Snapshots of equilibrated adsorbed state of fatty acid molecules on the \( m \)SiO\(_2\) surface at room temperature for different deprotonated degree, \( R \), increasing from 0.1 to 1.0. The dashed line is a reference line showing the height of the largest assembly at \( R = 0.1 \), \( D_{h,max} \). Functional groups (propyl ammonium ions) are drawn as grey licorice bonds. Bare silica is shown in grey solid van der Waals spheres. The coloring method of fatty acid molecules are the same as in Figure 4.2. Water and counterions are not shown here for clarity. The characteristic assembly is marked by the black arrow. At \( R = 0.1 \), a large fatty acid assembly with discrete negative charges is adsorbed on the positively charged \( m \)SiO\(_2\) surface. As \( R \) is increased, discrete small patches are formed instead of the large assembly on the \( m \)SiO\(_2\) surface, inducing a decrease of the mean height of surface aggregates, \( D_h \).

For the characteristic assembly, the probability distribution of the alkyl chain length (C\(_1\)-C\(_{10}\) distance) is shown in Figure 4.5a. Further, the density of all atoms and headgroups (oxygen in \(-COOH\) and \(-COO^-\) groups) as a function of the distance from the bare silica surface are shown in Figure 4.5b and 4.5c, respectively. To characterize the shape of the assembly quantitatively, we
Figure 4.5. (a) Probability distribution of the alkyl chain length of fatty acid molecules for various \( R \). The density of (b) all atoms and (c) headgroups (oxygen in \(-\text{COOH}\) and \(-\text{COO}^-\) groups) in the characteristic assembly as a function of the distance from bare silica surface for \( R \) values of 0.1, 0.3, 0.5 and 1.0. These \( R \) values feature the acidity of the solution from below to above its \( pK_a \). (d) Distribution of the angle between fatty acid molecule in the characteristic assembly and \( m\text{SiO}_2 \) (defined as the angle between \( C_{10}-C_1 \) vector and \( m\text{SiO}_2 \) surface normal) as a function of \( R \). All profiles are normalized in the way that the integration area is the same.

As shown in Figure 4.5a, the distribution of the alkyl chain length in the adsorbed characteristic assembly for different \( R \) is very similar to that in bulk water in Figure 4.2g, where
the distribution is trimodal, and most fatty acid molecules are in contracted configurations (peaks near 10.5 Å and 9.8 Å) rather than in fully extended configurations (peak at 11.3 Å). Thus, the assemblies are in fluid phase. Further, as illustrated in Figure 4.5b, the width of the distribution profile of all atoms in the characteristic assembly decreases with increasing $R$, suggesting that the size of the characteristic assembly drops, i.e. the fatty acids are more likely to spread over the $m$SiO$_2$ surface at a higher deprotonated degree due to the strong electrostatic attraction from the surface (see later discussions). The information on the shape of the characteristic assembly is demonstrated in Figure 4.5c and 4.5d together with the snapshots in Figure 4.4. We find that the morphology and phase behavior of adsorbed fatty acid assemblies at the $m$SiO$_2$-water interfaces show some similarity compared to that in bulk water at low and intermediate $R$ ($R \leq 0.5$) values, while at high $R$ ($R > 0.5$) there are considerable differences. The comparison is summarized in Table 4.2. At $R = 0.1$, the characteristic assembly is a bulky assembly comprising 4 deprotonated fatty acid molecules and 66 fatty acid molecules. This assembly is bound to the propyl ammonium groups on the $m$SiO$_2$ surface via carboxylate ions (red beads) with all hydrophobic alkyl chains (blue chains) aggregated. (Figure 4.4a) The density profile (Figure 4.5c) and the orientation distribution (Figure 4.5d) of the headgroups is ultra-broad, implying that fatty acid molecules in the assemble are disordered. This morphological behavior is a typical characteristic of the oil drop, and it is similar to its phase behavior in bulk water in Figure 4.2a.

As $R$ is increased to 0.5, the vertical density profile of the headgroups has two peaks at $z = 6.5$ Å and 22 Å (Figure 4.5c), demonstrating that the headgroups are concentrated at these two positions. This is an indicator of a bilayer-like structure, similar to the morphology in bulk water, (Figure 4.2d) which is further supported by the snapshot in Figure 4.4d. In addition, the angle distribution profile (Figure 4.5d) has a major peak at $\theta_z = 35^\circ$ with a minor broad peak near 135°.
The leaf peak represents the orientation of the bottom leaflet while the right peak stands for the top leaflet of the bilayer. The intensity of these two peaks competes, suggesting that the fatty acid molecules in the bilayer in Figure 4.4d have a preferential parallel/antiparallel arrangement, and the direction of the bilayer is 35° from the mSiO$_2$ surface normal.

Table 4.2. Comparison of the morphology and phase behavior of decanoic acid assemblies in bulk water and adsorbed at mSiO$_2$-water interfaces at 298 K.

<table>
<thead>
<tr>
<th>Acidity of fatty acid solution</th>
<th>Phase behavior in bulk water</th>
<th>Degree of deprotonation of fatty acid, $R = \frac{N_{\text{COO}^-}}{N_{\text{COOH}} + N_{\text{COO}^-}}$</th>
<th>mSiO$_2$-water interfacial phase behavior</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH &lt; $pK_a$</td>
<td>Oil droplets$^a$ 23,24,108</td>
<td>0.1</td>
<td>Oil droplet</td>
</tr>
<tr>
<td>pH $\cong pK_a$</td>
<td>Bilayers$^a$ or vesicles$^{23,28,108}$</td>
<td>0.5</td>
<td>Bilayer</td>
</tr>
<tr>
<td>pH $&gt; pK_a$</td>
<td>Aqueous spherical micelles$^{a,23,108}$</td>
<td>0.7</td>
<td>Bilayer</td>
</tr>
</tbody>
</table>

$^a$Phase behavior and morphology observed in Figure 4.2.

As $R$ is further increased above 0.5 ($R = 0.7$ and 1.0), the bilayer structure can still be observed (see Figures 4.4e and 4.4f), which differs from the spherical micellar structure in aqueous bulk solution in Figure 4.2e and 4.2f. Specially at $R = 1.0$ where all fatty acid molecules are deprotonated, the density profile of the headgroups has a ultra-sharp peak at $z = 6.5$ Å and a minor broad peak at $z = 20$ Å. (Figure 4.5c) The angle distribution of the C$_{10}$-C$_1$ vector has a major peak
near $\theta_z = 45^\circ$ with a tail extended to $180^\circ$ (Figure 4.5d) The sharp left peak in the density profile and the major peak in angle distribution implies that the bottom leaflet of the bilayer are adsorbed and aligned in the same direction due to the strong electrostatic attraction from $m$SiO$_2$ (see 4.3.3). In contrast, the molecules in the top leaflet are not always aligned in an ordered way, resulting in the broad second peak in Figure 4.5c and 4.5d.

In summary, the structure and phase behavior of decanoic acid assemblies can thus be altered by introducing an external charged surface. The bilayer-like structure can still be formed even though most of the decanoic acid molecules are deprotonated, and this is important for the preparation of a bilayer-type structure of fatty acids in a wider pH (deprotonated degree) range. The key to understanding the mechanism responsible for this type of morphological/phase behavior lies in understanding the external forces brought by the external surface.

### 4.5 Origin of Morphological Changes as a Function of Degree of Deprotonation

The spontaneous self-assembly and adsorption of fatty acid molecules are governed by (i) electrostatic attraction between the deprotonated fatty acid molecules (carboxylate ions) and $m$SiO$_2$ surface, and (ii) van der Waals interaction between the hydrocarbon tails of the neighboring fatty acid molecules. To understand the driving force that triggers the morphological change at different $R$, the electrostatic interaction$^{110}$ ($E_{elec}$) between fatty acid and $m$SiO$_2$ as well as the van der Waals interaction ($E_{vdW}$) among fatty acid alkyl chains are calculated and shown in Figure 4.6a and 4.6b, respectively, as a function of $R$. In addition, the contribution of electrostatic force from the $m$SiO$_2$ surface, $f_{elec}$ is also calculated by $f_{elec} = \frac{E_{elec}}{E_{elec} + E_{vdW}}$ in Figure 4.6c. It is clear that the contribution of electrostatic attraction from the $m$SiO$_2$ surface is amplified as the degree of deprotonation ($R$) is increased, while the contribution of van der Waals interaction attraction from the neighboring fatty acid molecules is diminished. The balance between these two forces provides
Figure 4.6. (a) Electrostatic interaction, $E_{elec}$ between fatty acid assemblies and $m$SiO$_2$ and (b) Van der Waals interaction, $E_{vdW}$ among fatty acid molecules as a function of $R$. (c) The contribution of electrostatic attraction to all attractions fatty acid gained, $f_{elec}$ at different $R$. $f_{elec}$ increases monotonically as a function of $R$, indicating the dominant role of FA-$m$SiO$_2$ attractive force over FA-FA van der Waals force in forming bilayer-like structures at a high deprotonated degree of fatty acid. Note: in figures, FA refers to fatty acids.

a solid explanation for the decrease in the size of the assembly in Figure 4.3b. For example, at $R = 0.1$, the strong van der Waals interaction among the fatty acid alkyl chains enables a higher degree of aggregation (Figure 4.3a). Meanwhile, the weak electrostatic interaction between the lowly charged assembly and the $m$SiO$_2$ surface immobilizes the large assembly on the substrate (Figure 4.4a and 4.5b).

At $R = 0.5$, the number of deprotonated fatty acid molecules is equal to that of protonated fatty acid molecules, and $f_{elec}$ is approximately 0.5 (see Figure 4.6b). The electrostatic interaction between fatty acid and $m$SiO$_2$ and the van der Waals attraction between the hydrocarbon chains of the molecules are of similar magnitude and compete. Compared to the assemblies formed in the region of $R < 0.4$, fatty acid assemblies at $R = 0.5$ have a lower aggregation number and more negative charges. For the characteristic assembly, the deprotonated fatty acid molecules act as
electrostatic linkers to mSiO$_2$ surface and form the bottom leaflet of the bilayer in Figure 4.4d, and the protonated fatty acids form the top leaflet above the bottom leaflet via strong vdW interaction.

As $R$ is increased to its maximum, all fatty acid molecules are present in their deprotonated (carboxylate) form. The contribution of electrostatic attraction from the mSiO$_2$ surface predominates over the van der Waals attraction from other fatty acid molecules (see Figure 4.6b). The relatively weak van der Waals interactions among the alkyl chains maintain the low aggregation number of all assemblies at $R = 1.0$ (see Figure 4.3a), and the strong attraction from mSiO$_2$ allows a large amount of deprotonated fatty acid molecules to be adsorbed via site-to-site binding. Further, in the characteristic assembly, the deprotonated fatty acid molecules can be bound to the mSiO$_2$ surface via strong electrostatic interaction, and they can form a packed monolayer (the bottom leaflet) by the lateral organization of the alkyl chains via van der Waals attraction, then the excess deprotonated fatty acid molecules are then packed to the monolayer to form a thin bilayer (see Figure 4.4f). The relatively weak vdW interactions decide that the top leaflet of the bilayer is not perfectly aligned in an ordered way, resulting in the weakening of the second peak in the orientation distribution in Figure 4.5d.

Therefore, the enhanced electrostatic interaction between fatty acid and mSiO$_2$, together with weakened van der Waals interaction between fatty acid molecules, drives the morphological transition of fatty acid assembly from large hydrophobic assembly (oil-in-water phase) to thin bilayers as a function of $R$. However, the van der Waals interaction cannot be weakened too aggressively to form an ordered bilayer structure. The observations on the morphological transition of fatty acid molecules on the mSiO$_2$ surface with increasing deprotonation agree well with our previous experimental results\textsuperscript{111}.
4.6 Summary

This chapter uncovered the role of two competing interactions and their correlation to the external experimentally adjustable parameter, pH for the formation of bilayer-like microstructure at the silica-water interface. This is achieved by strengthening the electrostatic binding attraction to the solid surface while slightly weakening the vdW interaction between neighboring fatty acid molecules. Based on this mechanism, the control of the formation of bilayer/vesicle structure can be achieved by adjusting the pH of the fatty acid solution to a wider range (pH > pK_a with R > 0.5), compared to the previous narrow range of pH ≅ pK_a with R = 0.5 in dilute alkaline solution. Further, the knowledge in those two competing interactions in deciding the solid-liquid interfacial phase behavior of fatty acid provided in this chapter also deliver insights into the application of hypothetical protocells.94–96, biosensors, enzyme-catalyzed reactions28, food, detergency, and the cosmetics industries, where the fatty acid assemblies are important structural components and need to be under control.
CHAPTER FIVE
THERMAL-RESPONSIVE ADSORPTION AND AGGREGATION OF ETHOXYLATED SURFACANT IN A CYLINDRICAL PORE§

5.1 Introduction

Adsorption and self-assembly of surfactants in porous materials have paved the way for bottom-up nanotechnology applications, such as surfactant flooding used for enhanced oil recovery, surfactant-enhanced water purification, surfactant-directed synthesis, and targeted drug delivery. The adsorption and self-assembly of surfactants to porous materials in the aqueous solution is governed by (i) the interaction between surfactant and pore-wall, (ii) the interaction among surfactant molecules, and (iii) the interaction between surfactant and water molecules. These interactions are determined by the chemical design of surfactant molecules and the pore material. Specifically, for ethoxylated surfactants in porous silica materials, the oxyethylene groups in surfactants and the hydroxide groups on the pore wall determine the key interaction being hydrogen bonding (H-bonding). H-bonding (Dn–H⋯Ac) is an intermolecular interaction between a polar group Dn–H acting as the bond donor and a more electronegative atom acting as the bond acceptor. The strength of a H-bond is determined by the electronegativity of the acceptor, and it can also be influenced by environmental properties such as local concentration, temperature, hydrophilicity, etc. that affect important properties such as adsorption. Although there has been a lot of experimental work on the adsorption and phase behavior if the ethoxylated surfactants, the underlying mechanisms regarding the H-bonding are still not well understood. Therefore, the goal of this chapter is to study the temperature response of H-bonding as a function

§Figure 5.2a-d and Figure 5.4a-c have been published in Ref. and are reproduced here with the permission from American Chemical Society.
of hydrophilicity of the surfactants, and in the next chapter, this investigation will focus on the thermal response of H-bonding as a function of pore size.

In bulk solution, the nonionic ethoxylated surfactants, n-alkyl poly(oxyethylene) ether surfactants (abbreviated as C\textsubscript{n}E\textsubscript{m}), are known to be water-soluble through H-bonding between the hydrophilic oxyethylene groups and water molecules. The total H-bonding interaction energy between C\textsubscript{n}E\textsubscript{m} and water molecules is known to be highly influenced by temperature. As reported by Bock and Gubbins,\textsuperscript{40} the surfactant-water H-bonding is weakened with increasing temperature. This reduction in H-bonding results in an increase in rotational entropy, i.e. the molecules rotate faster at higher temperatures, leading to the decrease of the surfactant’s solubility and phase separation above its cloudy temperature (see section 1.2.3). Compared to their phase behavior in bulk water, the adsorption and phase behavior of C\textsubscript{n}E\textsubscript{m} in silica pores are more complicated and harder to predict. In addition to the C\textsubscript{n}E\textsubscript{m}-water H-bonding in bulk solution (no adsorbents), the hydroxide groups (–OH) on silica can form silica-water H-bonds as well as silica-C\textsubscript{n}E\textsubscript{m} H-bonds. The temperature response of all three types H-bonds play important roles in determining the adsorption and phase behavior of C\textsubscript{n}E\textsubscript{m} molecules.

There has been considerable work focusing on the adsorption and phase behavior of C\textsubscript{n}E\textsubscript{m} surfactants in porous silica materials, but a full analysis regarding the mechanism, i.e., the responses of the three types of H-bonding mentioned above, are lacking. For example, small-angle neutron scattering (SANS) experiments have been performed to study the adsorption behavior of C\textsubscript{12}E\textsubscript{5} in a silica pore at room temperature near its CMC (see section 1.2.3).\textsuperscript{117–119} C\textsubscript{12}E\textsubscript{5} molecules first form patchy bilayers adsorbed on the silica pore walls. Later, a third molecule, lysine that forms stronger H-bonds with the silica pore walls, is added to the system where it is used to interrogate the H-bonding between silica and C\textsubscript{12}E\textsubscript{5}, leading to the detachment of C\textsubscript{12}E\textsubscript{5} assemblies.
However, an explicit H-bonding analysis cannot be easily obtained from the experiments. Computationally, mesoscale dissipative particle dynamics (DPD) simulations have been performed by Müter et al. to study the self-assembly of C\textsubscript{12}E\textsubscript{5} in cylindrical pores\textsuperscript{118,120,121}. They directly manipulate the interaction between the coarse-grained surfactant particles and pore wall by rescaling the non-bonded parameter of the pore wall, inducing the structural change of adsorbed surfactant assemblies at room temperature. In addition, they simulate the adsorption behavior of short-chain surfactant (C\textsubscript{6}E\textsubscript{3}) as a function of concentration, temperature and pore size (diameters of 10 nm, 35 nm and 50 nm) with an emphasis on the high concentration region. They found that the surfactants form a depletion region near the pore surface above the surface-azeotropic point, and the volume of the depletion region increases with the increasing pore size. These studies provide reliable information on the phase behavior of C\textsubscript{n}E\textsubscript{m} surfactants under different external parameters. However, the change of the H-bond interactions that give rise to these phenomena is not fully understood due to the challenge in tracking the number and strength of hydrogen bonds (atomic scale) in the mesoscale simulations where the surfactant molecules are coarse-grained.

In this chapter, we overcome those limitations by using all-atom molecular dynamics (MD) to study the temperature response of hydrogen bonding for C\textsubscript{6}E\textsubscript{m} molecules with different oxyethylene groups (different \(m\) value), namely, C\textsubscript{6}E\textsubscript{3} and C\textsubscript{6}E\textsubscript{5}. The temperature response of the total H-bond interaction energy (the nonbonded term in section 2.1.1) are shown to be responsible for the number change of these three types of H-bonds, namely, (i) silica-water H-bonds, (ii) silica–C\textsubscript{6}E\textsubscript{m} H-bonds, and (iii) C\textsubscript{6}E\textsubscript{m}-water H-bonds. It is found that for both systems, silica-C\textsubscript{6}E\textsubscript{m} H-bonding is independent of temperature and strong enough to allow for part of the surfactants to be strongly adsorbed on the pore wall and thereby, to serve as nucleation sites. However, silica-water and C\textsubscript{6}E\textsubscript{m}-water H-bond interactions decrease significantly as a function of temperature, allowing
for the aggregative adsorption of both C₆E₃ and C₆E₅ to the silica pore walls. Moreover, C₆E₃-water H-bonding is consistently weaker than C₆E₅-water H-bonding throughout the temperature region we studied, allowing C₆E₃ to be released from water more easily and to condense at the pore wall with increasing temperature. Consequently, the weaker C₆E₃-water H-bonding makes C₆E₃ assemblies to have a higher temperature sensitivity compared to the C₆E₅ assemblies.

5.2 Simulation Model and Setup

We simulate the adsorption and aggregation of surfactant C₆E₅ and C₆E₃ in a cylindrical silica pore of diameter 8.6 nm. To construct the pore structure, we first build an α-quartz silica supercell block by using the inorganic builder plugin in VMD¹²², then the cylindrical pore of diameter $D_p$ is carved from the silica block (Figure 5.2a). Next, O and Si edge atoms are passivated on internal pore walls as well as pore ends with hydrogen atoms and hydroxyl groups, resulting in a hydrophilic pore surface occupied by =Si(OH)₂ and ≡Si(OH) silanol groups, as shown in the inset of Figure 5.2a. The parameters for non-bonded and bonded interactions of silica are adopted from Heinz et al.⁵⁰ (see section 1.2.4 and 2.1), which are sufficient to describe silica’s bulk and surface properties, and their results were shown to be in excellent agreement with experimental measurements¹²³–¹²⁵.

The original structure of C₆E₅ and C₆E₃ monomer is constructed via the Automatic Topology Builder¹²⁶ as illustrated in Figure 5.1b. The parameters for non-bonded and bonded interactions are taken from the Charmm database¹²⁷ for linear and cyclic ethers. The TIP3P model is used to represent the water model¹⁰¹. C₆E₅ and C₆E₃ monomers share the same alkyl chain and differ only in the number of oxyethylene groups (−OCH₂CH₂). The number of oxyethylene groups not only determines the hydrophilicity of the surfactant monomer but also plays a role in determining the structure and the thermal responsive properties of the self-assemblies. Given that
the CMC (section 1.2.3) of C₆E₅ and C₆E₃ is near 0.1 M, a concentration of 0.3 M is used in this study to ensure that enough molecules are provided to form micelles.

Figure 5.1. (a) Silica cylindrical pore model, where $D_p$ represents the diameter of cylindrical pore. The inset next to (a) is the chemical sketch of the functionalized hydrophilic pore wall. Silicon, carbon, oxygen and hydrogen atom are colored in yellow, grey, red and blue, respectively. (b) Topology of C₆E₃ and C₆E₅ monomers. (c) Sideview of the simulation box for pore-surfactant system. In (d), the pore region is open-ended and connected to bulk region, which serves as a reservoir of C₆Em molecules.

After the preparation of the silica pore and surfactant molecules, we embed the silica pore in a large simulation box with the ends of the pore is open to the ‘bulk’ region, as shown in Figure 5.2d. The simulation box is then filled with a 0.3 M solution of C₆E₅/C₆E₃, thereby keeping the
concentration of surfactant solution the same both in and out of the pore region with the region at the end of the pore serving as a surfactant reservoir.

Energy minimization of all systems are performed using the steepest decent algorithm with no temperature assigned to generate starting configurations with appropriate bond distances. Given that the lower critical solution temperature (LCST) of C₆E₃ is near 45 °C, (also see section 1.2.3), the simulations are performed at 4 different temperatures up to the LCST before the phase separation occurs, namely, 20 °C, 30 °C, 38 °C, and 45 °C. The simulation starts with the lowest temperature and ending with the highest temperature where the initial configuration is obtained from the neighboring low temperature equilibrated final structure (except for the lowest temperature). At each temperature point, the pre-equilibration simulation is conducted in two steps: (i) Equilibration in an NVT ensemble (see section 2.3.1) to stabilize the temperature by using V-rescale thermostat; (ii) Equilibration in an NPT ensemble (constant number of atoms, pressure, and temperature) to initiate the system to 1 bar using Berendsen barostat, which has been considered as the most efficient way to scale a box at the beginning of a run. After the simulation box reaches the desired temperature and pressure, a 10-ns run is then performed in the NPT ensemble using the V-rescale thermostat for temperature coupling and the accurate Parrinello-Rahman barostat. The equilibration of surfactant assemblies is determined through the convergence of their RMSD referenced to their initial configurations (see section 2.3.2). The dynamical equations are solved using a leap-frog integration algorithm with a timestep of 1 fs is used for all equilibration simulations. The particle mesh Ewald (PME) method is used for long-range Electrostatic interaction (charge-charge interaction) using 4th order interpolation. The cutoff for non-bonded Lennard-Jones interaction and the real-space of Ewald sum is 1.1 nm (given that
\( \sigma \) for O atom in LJ potential in equation 2.3 is 0.36 nm where the cutoff is usually set to 3 times of \( \sigma \).

**5.3 Aggregative Adsorption of \( C_6E_3 \) and \( C_6E_5 \) in a Cylindrical Pore**

By introducing a cylindrical silica pore, the aggregation behavior of surfactant molecules will be more complex and difficult to predict than that in bulk due to additional interactions between surfactant and the pore wall. It is believed that the dominant binding mechanism of \( C_nE_m \) surfactants on a hydrophilic surface is due to hydrogen bonding between oxyethylene (EO) group and hydroxy group on the substrate.\(^{129-132} \) However, both EO groups and hydroxy groups are strongly hydrated in water, thereby reducing the net interaction between surfactant molecules and the solid surface. By increasing the temperature, both solid surface and EO groups can be dehydrated, which is beneficial to the adsorption. To determine the role of hydrogen bonding in the temperature-responsive adsorption behavior of \( C_nE_m \) surfactants with different EO groups, we compare the temperature-induced morphological transformation of \( C_6E_3 \) assemblies with that of \( C_6E_5 \). The snapshots of the equilibrated \( C_6E_3 \) and \( C_6E_5 \) assemblies in a silica pore is displayed in Figure 5.2 as a function of temperature, and only the ones adsorbed in the pore are shown for better visualization. We found that at 20 °C, \( C_6E_3 \) molecules are adsorbed on the pore surface as individual molecules and several small patches all around the pore wall (panel a), while \( C_6E_5 \) molecules are adsorbed as both discrete micelles and small amorphous clusters (panel e). As the temperature is increased, both pre-adsorbed \( C_6E_3 \) and \( C_6E_5 \) assemblies grow to larger aggregates when un-adsorbed or weakly adsorbed surfactant molecules/clusters coalesce with strongly adsorbed patches, but with different morphologies. At 38 °C, a fragmented \( C_6E_3 \) bilayer is formed, as shown in Figure 5.3c, and \( C_6E_5 \) assemblies are adsorbed as globular micelles. As the temperature
is further increased to 45 °C, the $C_6E_3$ bilayer transforms to a globular micelle, while $C_6E_5$ assemblies do not show any significant difference.

Figure 5.2. Snapshots for equilibrated (a-d) $C_6E_3$ and (e-h) $C_6E_5$ in the silica pore as a function of temperature in the projection along the pore’s central axis. All atoms contained in silica framework is displayed as grey stick. For surfactant molecules, all CH$_2$ groups in hydrophobic tails are shown in green beads, and CH$_2$, O, OH groups in hydrophilic heads are shown in red beads. Only pore-adsorbed surfactant molecules and assemblies are shown here for better visualization, and water molecules are not shown here for clarity. Depth cue is applied along the central axis.

To characterize the size of the adsorbed surfactant assembly, we count the aggregation number (the number of surfactant monomers present in each assembly) as a function of temperature. As not every assembly is growing at the same time, we choose the largest one as the characteristic assembly to better capture the growth of the assembly as a function of temperature. We plot the characteristic assembly’s aggregation number, $N_a$, as a function of temperature in Figure 5.3a. We find that as the temperature is increased, $N_a$ for $C_6E_3$ has an immediate sharp increase, while $N_a$ for $C_6E_5$ grows more slowly. Thus, $C_6E_3$ molecules are more sensitive to
temperature than C₆E₅ in a cylindrical silica pore. The growth of the assembly on the pore wall is in agreement with the known aggregative adsorption of C₆E₅₃ surfactants on hydrophilic surfaces.⁴⁰

Figure 5.3. (a) Aggregation number of the characteristic assembly, \(N_a\) and (b) Total surface area of surfactant molecules/assemblies both in bulk solution and adsorbed at pore wall, \(A_t\) as a function of temperature. The Connolly surfaces of (i) a single C₆E₃ molecule at 20 °C, (ii) an adsorbed C₆E₃ assembly at 45 °C and (iii) an adsorbed C₆E₅ assembly at 20 °C are inserted in (b). The inserted Connolly surfaces schematically show that \(A_t\) decreases when surfactants are present as larger assemblies in simulation box at an elevated temperature.

To show that the growth of primary micelle at the elevated temperature is not incidental, the size of all assemblies in the simulation box is characterized by calculating their solvent-accessible surface area via the double cubic lattice method¹. In the method, solvent probes (spheres of radius 0.14 nm) are used to roll along the envelope of the van der Waals surface of a single surfactant molecule or an assembly, and the surface area is then calculated through multiplying the number of probe spheres consumed by the portion of surface area each sphere represents. The surface of a molecule/an assembly covered by probe spheres is referred to as a Connolly surface, shown as the insets in Figure 5.3b. The total surface area of all assemblies in the simulation box, \(A_t\), is plotted as a function of temperature in Figure 5.3b. As can be seen in Figure 5.3b, the Connolly surfaces show that \(A_t\) decrease when the size of assembly increases at an
elevated temperature, where (i) represents a single C₆E₃ molecule at 20 °C, (ii) is an adsorbed C₆E₃ assembly at 45 °C, and (iii) is an adsorbed C₆E₅ assembly at 20 °C. It is noted that for both C₆E₃ and C₆E₅ system, $A_t$ decreases as a function of temperature even though the total number of surfactant molecules in each simulation box is fixed. This can be understood from the growth of the assembly where $A_t$ decreases due to more molecules being encapsulated inside the assembly, as evidenced by the Connolly surface in the insets. We also observe that $A_t$ has a dramatic drop for C₆E₃ assemblies at 30 °C, while for C₆E₅, $A_t$ decreases slowly for all temperatures within the temperature range. The difference in the slope of $A_t$ as a function of temperature is a further evidence that C₆E₃ is more sensitive to temperature than C₆E₅. $A_t$ provides a good qualitative understanding that C₆El surfactants with fewer EO groups are more responsive to temperature both in bulk solution and adsorbed at hydrophilic surfaces.

5.4 Origin of the Growth of the Assembly

Based on the analysis of the size of adsorbed C₆E₃ and C₆E₅ assembly in a pore as a function of temperature, we believe the C₆E₃ assembly adsorbed in a silica pore is more temperature-responsive due to its lower hydrophilicity. Although the resulting morphology and temperature sensitivity of adsorbed C₆E₃ and C₆E₅ assemblies are different, the mechanism behind the temperature-induced morphological change is similar, and we believe it is related to the temperature response of hydrogen bonding occurring in the silica pore/C₆El/water systems. The three types of hydrogen bonds shown schematically in Figure 5.4a are:

**Type I** – Silica-water hydrogen bond i.e. HO-H$_{\text{water}}$---O$_{\text{silica}}$ and SiO-H$_{\text{silica}}$---O$_{\text{water}}$;

**Type II** – Direct hydrogen bond between silica and surfactant i.e. SiO-H$_{\text{silica}}$---O$_{\text{surfactant}}$ and O$_{\text{surfactant}}$-H$_{\text{surfactant}}$---O$_{\text{silica}}$;
**Type III** – Hydrogen bond between surfactant and water i.e. HO-H\textsubscript{water}---O\textsubscript{surfactant}, C-H\textsubscript{surfactant}---O\textsubscript{water}\textsuperscript{134} and O\textsubscript{surfactant}-H\textsubscript{surfactant}---O\textsubscript{water}.

We track the number of hydrogen bonds for different H-bond types as a function of temperature (Figure 5.4b-d). The GROMACS\textsuperscript{73} package was used to calculate the number of H-bonds using a grid search method\textsuperscript{135}, where the simulation box is decomposed into small grids, and H-bonds are searched within each grid. The bond length of H-bond varies from 1.5 to 3.0 Å.\textsuperscript{35} Given that, the cutoff length is set to be 3 Å, and the cutoff angle is 20°. It is noted that the process of finding H-bonds is to check if the angle and distance between the electron donor and the accept meet the criteria, thus results can be different by using different analysis packages during the dynamic equilibrium process. We only focus on the number change rather than the absolute value of H-bond as a function of temperature, thus any package could be used.

Furthermore, to examine the contribution of electrostatic term (Ewald term) and the van der Waals term (LJ term) to the hydrogen bonding in the MD simulation, the electrostatic interaction, \(E_{elc}\) and the van der Waals interaction, \(E_{vdW}\) for all above pairs are extracted and plotted in Figure B.1. It shows that both electrostatic and van der Waals interaction contributes to the H-bonding, while the electrostatic term is consistently higher than the vdW term (about 1 to 2 times higher). Thus, the H-bond interaction energy is predominantly electrostatic, as reported in the Charmm forcefield we applied\textsuperscript{98,136}. The H-bond interaction energy as a function of temperature is plotted in Figure 5.5e-g.

The silica-C\textsubscript{6}E\textsubscript{m} H-bonding (Type II) is nearly independent of temperature between 30 °C and 45 °C in Figure 5.4c and 5.4f, revealing that part of the surfactant molecules/patches are strongly adsorbed at the pore wall. As temperature is increased from 20 °C to 30 °C, the H-bond interaction energy between silica and C\textsubscript{6}E\textsubscript{m} \((U_{SE})\) decreases, inducing the breakage of the H-bonds.
Figure 5.4. (a) Schematic representations of three pair types of hydrogen bond involved in the process of aggregative adsorption. Color codes of atoms can be found in Figure 5.1, and the H-bonds are represented by green dashed lines. Number of hydrogen bond between (b) silica and water, (c) silica and C$_6$E$_m$, and (d) C$_6$E$_m$ and water as a function of temperature. H-bond interaction energy between (e) silica and water, (f) silica and C$_6$E$_m$, and (g) C$_6$E$_m$ and water as a function of temperature. Each number is averaged along the last 1-ns equilibrium MD trajectory, and the error bars show the standard deviation, indicating a minor fluctuation during the dynamic equilibrium.
between the silica surface and weakly-adsorbed $C_{6}E_{m}$ molecules/patches. Thus, the weakly-adsorbed $C_{6}E_{m}$ molecules found at 20 °C detach from the surface and migrate to coalesce with the strongly bound patches, as shown in Figure 5.2a-b and e-f. In Figure 5.4e and 5.4g, the H-bond interaction energy between silica and water as well as that between $C_{6}E_{m}$ and water decreases significantly with increasing temperature, leading to the drop in the number of Type I and Type III H-bonds in Figure 5.4b and 5.4d. The breakage of silica-H$_2$O and $C_{6}E_{m}$-H$_2$O H-bonds indicate that the bound water molecules are released from both the silica surface and $C_{6}E_{m}$ molecules, dehydrating both silica surface and $C_{6}E_{m}$ molecules/patches. In this regard, the $C_{6}E_{m}$ molecules/patches in the pore can aggregate on the pore surface via hydrophobic effect$^{37,38}$ (see section 1.2.3). The above analysis indicates that the temperature response of the hydrogen bonding induces the aggregative adsorption of both $C_{6}E_{3}$ and $C_{6}E_{5}$ surfactants. During the temperature-responsive aggregative adsorption process, the temperature-independence of the silica-$C_{6}E_{m}$ H-bonding allows the strongly adsorbed $C_{6}E_{m}$ molecules/patches to act as nucleation sites, and the decline of silica-H$_2$O and $C_{6}E_{m}$-H$_2$O H-bonding leads to the subsequent condensation of surfactant assemblies to the pore wall.

Apart from the similarity in the temperature response of H-bonding, the H-bond interaction energy between silica and $C_{6}E_{3}$ (Figure 5.4f) is consistently higher than that between silica and $C_{6}E_{5}$, indicating that $C_{6}E_{3}$ molecules are more preferentially adsorbed on the silica pore via direct $\text{Si-OH}_{\text{silica}}-\text{O}_{\text{surfactant}}$ and $\text{OH}_{\text{surfactant}}-\text{O}_{\text{silica}}$ bonding compared to $C_{6}E_{5}$, as displayed in Figure 5.2. The $C_{6}E_{3}$-H$_2$O H-bonding is steadily weaker than the $C_{6}E_{5}$-H$_2$O H-bonding (Figure 5.4 d and 5.4g), making it is easier to break the $C_{6}E_{3}$-H$_2$O H-bonds and to release $C_{6}E_{3}$ from the H-bond network of the bulk water with increasing temperature. This feature leads to the higher temperature sensitivity of $C_{6}E_{3}$ compared to $C_{6}E_{5}$ (Figure 5.3).
5.5 Summary

In this chapter, we probe the temperature response of the H-bond interaction energy as a function of temperature, and demonstrate the role of H-bonding in determining the similarity in the aggregative adsorption of $C_6E_3$ and $C_6E_5$ molecules in a hydrophilic cylindrical pore (diameter of 8.6 nm) at elevated temperatures as well as the difference in temperature sensitivity of the adsorbed $C_6E_3$ and $C_6E_5$ assemblies. For the similarity part, the aggregative adsorption is initiated by the same temperature response of H-bonding between (i) silica surface and water, (ii) silica surface and $C_6E_m$, and (iii) $C_6E_m$ and water. The temperature-independence of silica surface-$C_6E_m$ H-bonding enables the strongly adsorbed $C_6E_m$ molecules/patches to serve as the nucleation sites, and the temperature-triggered breakage of silica-water and $C_6E_m$-water H-bonds lead to their condensation via hydrophobic effects.

On the other hand, compared to $C_6E_5$, the weaker $C_6E_3$-water H-bonding increases the likelihood of bond breakage with increasing temperature, leading to higher sensitivity and faster response to temperature. Thus, parameters that influence the H-bonding associated with $C_nE_m$ molecules and hydrophilic adsorbents can be used to program the morphology and thermal property of $C_nE_m$ assemblies on hydrophilic surfaces. For example, the thermal sensitivity of the adsorbed can be decreased by increasing the ethoxylation degree ($m$ value) of $C_nE_m$ molecules. Molecules that can form stronger H-bonds with silica surface can be used to induce the desorption of the ethoxylated surfactant assemblies. This finding will provide the experimentalists with insights in choosing the right parameters and procedures to guide the adsorption and self-assembly of nonionic surfactants with desired growth/desorption rate and structures.
CHAPTER SIX
EFFECT OF PORE CONFINEMENT ON THERMAL RESPONSIVENESS OF SURFACTANT ASSEMBLIES

6.1 Introduction

In Chapter 5, we compared the thermal response of H-bonding in the silica pore/C₆E₃/water system with that in the silica pore/C₆E₅/water system. For both C₆E₃ and C₆E₅, the silica-surfactant H-bonds is responsible for the initial adsorption and the initial formation of adsorbed patches that serve as nucleation sites for subsequent condensation. Further, both C₆E₃-water and C₆E₅-water H-bond interactions decrease in response to temperature, driving the subsequent surfactant assemblies to be released from water and to aggregate with the pre-adsorbed surfactants. The difference in the number of oxyethylene groups in C₆E₃ and C₆E₅ has a significant influence on the temperature response of C₆E₃-water H-bonding, where the weaker C₆E₃-water H-bonding has a more instantaneous response to temperature, leading to a higher temperature sensitivity of C₆E₃ assemblies compared to C₆E₅ assemblies.

The thermal response is not only dependent on the number of oxyethylene groups in the surfactants but also on the pore confinement. Thus, in this chapter, we focus on the thermal response of H-bond interactions in silica pore/C₆E₅/water system as a function of the cylindrical pore confinement (defined as the reciprocal of the cylinder radius), which leads to different thermal responsiveness of the assemblies in pores of different sizes. As the silica pore is immersed in the surfactant solution, and the surfactant molecules exist both in and out of the pore, the initial step of the adsorption process can always occur via strong H-bond interaction between the pore wall and surfactants within the pore regardless of the pore size. We believe that the change of pore size mainly affects the later stage of the aggregation process, where the assembly outside of the pore migrate into the pore and coalesce with the assemblies in the pore at a higher temperature. We
found that the decrease of C₆E₅-water H-bonding together with the increase of van der Waals interaction between the assembly in and out of the pore as a function of temperature drives the hydrophobic association of these two assemblies. The thermal responses of these two interactions are more hysteretic as the degree of pore confinement is increased, resulting in the shift of the coalesce temperature to a higher value in a pore of a smaller size. In other words, the surfactant assembly outside of a narrower pore is less sensitive to temperature than one that exits in a larger pore. Based on this knowledge, the temperature sensitivity and the transport rate of the ethoxylated surfactant assemblies can be tuned by the degree of pore confinement.

6.2 Simulation Model and Setup

In this study, we will simulate the temperature-induced adsorption and aggregation of surfactant C₆E₅ in cylindrical silica pores as a function of pore diameter (D_p), namely 8.6 nm, 4 nm, and 2 nm. The 8.6 nm pore structure is adopted from Chapter 5. The pore structure of size 4 nm and 2 nm is constructed following the procedure used in section 5.2 with the topologies shown in Figure 6.1 (b) and (c). It needs to be noted that the size of the smallest pore in this study was chosen to be about the size of a single C₆E₅ molecule in order to explore extreme condition. The degree of pore confinement can be defined by the parameter K₁, which is the reciprocal of the cylinder radius (K₁ = 1/Dₚ/2). The corresponding values of K₁ for pore diameters of 8.6 nm, 4 nm, and 2 nm are 0.23, 0.5, and 1, respectively. As a reference, self-assembly of surfactants in aqueous bulk solution is also simulated in a fixed rectangular box where the K₁ value is zero. As shown in Figure 6.1d, the simulation boxes of the silica pore/C₆E₅/water systems, where a fixed concentration (0.3 M) of C₆E₅ solution is placed both in and out of the pore. The simulations for all systems are conducted using ten temperature points starting at 20 °C and ending at 80 °C with the final configuration of the former temperature point being used as the starting structure for the
simulation at the next higher temperature. The equilibration process for each temperature point uses the same procedure described in section 5.2.

Figure 6.1. Cross-sectional view of silica pore model of diameter (a) 8.6 nm, (b) 4 nm and (c) 2 nm, respectively. The corresponding degree of pore confinement, $K_1$, for pore diameters of 8.6 nm, 4 nm and 2 nm is 0.23, 0.5 and 1, respectively. All silica pore models are placed in the periodic simulation box in x, y and z directions. (d) Side view of the simulation box for silica pore/C₆E₅/water system. The concentration of C₆E₅ solution in and out of the pore is the same. The simulation box is periodic in x, y and z directions, and it is drawn by black lines. (e) Temperature program of the heating process for all silica pore/C₆E₅/water systems. The heating and equilibration processes are conducted in a serial way, where the final configuration at the former temperature is used as the starting point for the simulation at the subsequent temperature.
6.3 Effect of Pore Confinement on the Coalescence Process

Figure 6.2. Snapshots of equilibrated states of C₆E₅ assembles in aqueous bulk solution (panel a1-a10), under pore confinement of $K_1 = 0.23$ (panel b1-b10), $K_1 = 0.5$ (panel c1-c10) and $K_1 = 1.0$ (panel d1-d10) as a function of temperature. The coloring method follows that in Figure 5.3. In all these systems, only aggregates are shown, while surfactant monomers and water molecules are not shown here for clarity. In panel b to e, the periodic simulation box is colored in black.

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To compare the thermal response of C₆E₅ assemblies as a function of K₁, the side view of the periodic boxes for all systems at the equilibrated states are displayed in Figure 6.2. The horizontal axis represents the increasing degree of confinement, and the vertical axis shows the temperature. At \( T < 60^\circ C \), the C₆E₅ assemblies are distributed both in and out of the silica pore at all \( K_1 \) values, while at \( T \geq 60^\circ C \), the assemblies located outside of the pore migrates into the pore and coalesces with the pre-adsorbed assembly in the pore regardless of the pore size. The assemblies aggregate at a higher temperature at all \( K_1 \) values, however, the exact temperature where the assembly out of the pore coalescence with the one in the pore does vary as a function of confinement.

To characterize the temperature-induced coalescence process, we extracted the number of aggregates, \( N \) in the simulation box, and the radius of gyration of all aggregates, \( R_g \) as a function of temperature for different \( K_1 \), as shown in Figure 6.3a and 6.3b, respectively. The radius of gyration for all aggregates distributed in the simulation box can be calculated via equation 6.1 as follows:

\[
R_g = \left[ \frac{\sum_i \| \mathbf{r}_i \|^2 m_i}{\sum_i m_i} \right]^{1/2} \tag{6.1}
\]

where \( m_i \) is the mass of atom \( i \), and \( \mathbf{r}_i \) is the position of atom \( i \) with respect to the center of mass of all aggregates in the simulation box. \( R_g \) can be used to characterize the mass distribution of the dispersed aggregates and the compactness of an aggregate.\(^{138-141}\) The decrease of \( N \) in Figure 6.3a and \( R_g \) in Figure 6.3b as a function of temperature indicates the aggregation of distributed patches followed by coalescence into one assembly. As shown in Figure 6.2, at \( K_1 = 0 \) (no pore confinement), two C₆E₅ micelles are formed at 20 °C (panel a1), and after raising the temperature
Figure 6.3. (a) Number of C₆E₅ aggregates assembly in the simulation box, N as a function of temperature under different degrees of confinement, \( K_1 \). (b) Radius of gyration of all assemblies distributed in the simulation box, \( R_g \) as a function of temperature for different \( K_1 \). The decrease of \( N \) to one together with the convergence of \( R_g \) as a function of temperature indicates that the distributed patches aggregate to one assembly, ultimately. (c) The sideview of the simulation box right before and after the coalescence between the assembly in and out of the pore. The temperature right after the coalescence, defined as \( T_c \), shifts to a higher value as the degree of confinement, \( K_1 \) is increased, revealing the decrease of temperature sensitivity of C₆E₅ assembly under extreme confinement.
to 30 °C, two adjacent micelles coalesce to form a large micelle (panel a2), inducing the sharp decrease of $R_g$ and the rapid convergence of $R_g$. As can be seen, increasing pore confinement slows down the aggregation process. At $K_1 = 0.23$, both $N$ and $R_g$ show a two-stage change as a function of temperature (see Figure 6.3a and 6.3b). In the region $20 °C < T \leq 45 °C$, the slow decline of $R_g$ is due to the coalescence of assemblies in the pore space. The large pore space allows for the migration and the increase of aggregation of surface-adsorbed patches leading to even larger patches. As $T$ is increased from 45 °C to 50 °C and at $K_1 = 0.23$, $R_g$ shows a sudden drop, and a large assembly located out of the pore (panel b4) now enters the pore and coalesces with the adsorbed assembly in the pore (panel b5). Snapshots right before and after the coalescence process are shown in Figure 6.3c for different $K_1$ values, and the temperature right after coalescence is defined as $T_c$. For example, $T_c$ for $K_1 = 0.23$ is equal to 50 °C. The coalescence of the two assemblies in and out of the pore is shown to be driven by the decreased $C_6E_5$-$H_2O$ H-bonding ($U_{EH}$) together with the increased van der Waals interaction ($U_{vdw}$) between the hydrophobic cores of these two assemblies. (see Figure 6.5d and 6.5e) The decreased $U_{EH}$ drives the $C_6E_5$ to escape from the H-bond network of water, and the sudden increase of the $U_{vdw}$ at $T_c$ leads to the hydrophobic association of these two assemblies.

At $K_1 = 0.5$, unlike the case of the larger pore ($K_1 = 0.23$) where four assemblies are dispersed and adsorbed on the inner pore wall (panel b1 in Figure 6.2), there are only two assemblies adsorbed in the pore (panel c1), the smaller number of adsorbed assemblies is due to the smaller pore space. Similarly, the assembly at $K_1 = 0.5$ also shows a two-step growth, whereas $T$ is increased from 20 °C to 38 °C the two adsorbed patches in the pore aggregate. In the range of $38 °C < T \leq 50 °C$, the two assemblies located in and out of the pore are approaching each other (panel c3, c4, and c5), resulting in a gradual decrease of $R_g$ in Figure 6.3b. Finally, the two
assemblies in panel c5 meet each other and aggregate into a large compact micelle (panel c6) that is driven by the decrease of $U_{EH}$ and the sudden increase of $U_{vdw}$ (Figure 6.5), leading to the convergence of $R_g$ at $T_c = 55 \, ^\circ C$.

At $K_1 = 1.0$, only one cluster is located in the pore with another cluster outside of the pore at 20 °C. (panel d1) In this circumstance, only one-step coalescence is observed. Unlike the assembly in the larger pore, the assembly in this narrow pore can only occur along the axial direction of the cylindrical pore due to geometric confinement. $U_{EH}$ decreases and $U_{vdw}$ develops a sudden increase at $T_c = 60 \, ^\circ C$, leading the two assemblies in and out of the pore (panel d6) to aggregate at 60 °C (panel d7).

In summary, $U_{EH}$ decreases as a function of temperature for all confinement geometries, indicating that C$_6$E$_5$ assemblies are dehydrating, leading the assemblies to an escape from water. Meanwhile, for higher $K_1$, the van der Waals interaction between the two assemblies in and out of the pore ($U_{vdw}$) demonstrates a slower increase in response to temperature, leading to a slower coalescence process of the assemblies (slower convergence of $R_g$) at a higher $K_1$. Thus, $T_c$ increases monotonically as a function of $K_1$, which clearly shows the decrease of temperature sensitivity of C$_6$E$_5$ assembly under extreme confinement.

### 6.4 Effect of Pore Confinement on the Shape of the Adsorbed Assembly

After the coalescence of the assemblies inside and outside of the pore at $T_c$, the shape of the assembly changes as a function of confinement ($K_1$) for increasing temperature above their $T_c$ values. For this section we will focus on the shape change of the assembly which is formed by the coalescence of the two assemblies in and out of the pore mentioned above. The relative shape anisotropy of a single assembly, $K^2$, can be calculated via equation 6.2, where $\lambda_x$, $\lambda_y$, and $\lambda_z$ are the principal moments of the inertia tensor of the assembly as shown in equation 6.3. $r_{i,x}$, $r_{i,y}$, and
$r_{i,x}$ is the distance between the position of atom $i$ and the center of mass of the assembly along the principal axes, $x$, $y$ and $z$, respectively.\textsuperscript{142,143}

$$K^2 = \frac{3}{2} \left( \frac{\lambda_1^4 + \lambda_2^4 + \lambda_3^4}{(\lambda_1^2 + \lambda_2^2 + \lambda_3^2)^2} \right) - \frac{1}{2}$$ \hfill (6.2)

$$\lambda_x = \left[ \frac{1}{2} \left( \frac{\sum (r_{i,x}^4 + r_{i,z}^4)m_i}{\sum m_i} \right) \right]^{1/2}, \quad \lambda_y = \left[ \frac{1}{2} \left( \frac{\sum (r_{i,y}^4 + r_{i,z}^4)m_i}{\sum m_i} \right) \right]^{1/2}, \quad \lambda_z = \left[ \frac{1}{2} \left( \frac{\sum (r_{i,x}^4 + r_{i,y}^4)m_i}{\sum m_i} \right) \right]^{1/2}$$ \hfill (6.3)

An assembly with ideal spherical symmetry has a $K^2 = 0$, and as $K^2$ increases above zero, the shape of the assembly will transform to an ellipsoid-like or a rod-like shape. As a reference, a rod-like C$_{12}$E$_5$ micelle comprising 120 monomers has a $K^2$ value of 0.08.\textsuperscript{143} To further characterize a micellar assembly, the ratio of exposed surface area of hydrophobic tails to the total surface area, $R_{\text{pho}} = \frac{A_{\text{alkyl tails}}}{A_t}$, is calculated. (see section 5.3.1) A globular micelle should have a low value of $R_{\text{pho}}$, as the hydrophobic tails are encapsulated into the core of the micelle. The $K^2$ value and $R_{\text{pho}}$ of the assembly after the coalescence in the range of $T_c \leq T \leq 80$ °C for different degrees of confinement is plotted in Figure 6.4a and 6.4b, respectively. At $K_1 = 0.23$, the exposed area ratio of hydrophobic tails ($R_{\text{pho}}$) is close to 0.1 throughout the temperature range of $T_c \leq T \leq 80$ °C, indicating that a micelle is formed in the largest pore. The $K^2$ value at $K_1 = 0.23$ first increases to 0.015 then goes back to 0 as a function of temperature, suggesting that the micelle first transforms to the “rod-like” conformation then goes back to the “spherical” shape. This morphological transition is driven by the change of silica-C$_6$E$_5$ H-bond interaction ($U_{SE}$), where the attraction is only from one side of the pore wall where it first increases then decreases (Figure 6.5c). At $K_1 = 0.5$ and 1.0, $U_{SE}$ fluctuates with temperature, as the adsorbed assembly can also feel the attraction from the opposite pore wall within the narrow pore. The fluctuation of $U_{SE}$ leads to the variation...
Figure 6.4 (a) Relative shape anisotropy, $K^2$ and (b) Ratio of exposed hydrophobic area, $R_{pho}$ of the C$_6$E$_5$ assembly as a function of temperature in the range of $T_c \leq T \leq 80$ °C for different degrees of confinement. Both $K^2$ and $R_{pho}$ at each temperature is averaged along the last 1-ns of the equilibrium trajectory, and the error bar is the standard deviation. As the degree of confinement ($K_1$) is increased, both $K^2$ and $R_{pho}$ are increased. It indicates that C$_6$E$_5$ surfactants can form a spherical micelle in a larger pore with low confinement, while they form a rod-like cluster in an ultra-small pore with extreme confinement, implying the effect of geometric confinement on the shape of the C$_6$E$_5$ assembly.

of the $K^2$ and $R_{pho}$ values of the assembly adsorbed in narrow pores, and the variation at $K_1 = 1.0$ is more significant compared to $K_1 = 0.5$. At $K_1 = 0.5$, $U_{SE}$ has a slight increase as $T$ is increased from 55 °C to 60 °C, and the increased attraction comes from all sides of the pore wall. This attraction from all sides of the pore walls drives the micelle to transform from an ellipsoid-like to a spherical shape, resulting in a notable decline of both $K^2$ and $R_{pho}$. This morphological transition can be observed in panel c6 and c7 of Figure 6.2. As the degree of confinement is further increased to 1.0, both $K^2$ and $R_{pho}$ are remarkably higher than those with a low degree of confinement. This feature can be captured in Figure 6.2, where the assembly at $K_1 = 1.0$ is not a micelle but rather a rod-like cluster. Moreover, the cluster’s relative shape anisotropy first increases sharply and then diminishes as a function of temperature with large fluctuations marked by the error bar. This is induced by the large fluctuation of $U_{SE}$ at $K_1 = 1.0$, where the attraction from the surrounding pore.
wall drives the cluster to adjust its configuration to fit in the ultra-narrow pore channel. Based on the above analysis, it can be concluded that the geometric confinement of the cylindrical pore has an incontrovertible impact on the shape of the adsorbed assembly above $T_c$ originating from the silica-C$_6$E$_5$ H-bond interaction. The surfactants form a spherical micelle in the larger pore (low degree of confinement), while they form a rod-like cluster in an ultra-narrow pore whose size is approximately equal to the size of the C$_6$E$_5$ monomer (extreme confinement).

6.5 Analysis of Hydrogen Bonds and Energetics

We believe that the shift of $T_c$ as a function of the degree of confinement ($K_1$) is related to the temperature response of hydrogen bonding in the silica pore/C$_6$E$_5$/water system and the interaction between two assemblies. For the H-bonding part, we tracked the number change of silica-C$_6$E$_5$ H-bonds ($N_{SE}$) and C$_6$E$_5$-H$_2$O H-bonds ($N_{EH}$) as a function of temperature for different $K_1$, as shown in Figure 6.5a and 6.5b, respectively. The contributions of electrostatic term, $E_{elec}$ and the van der Waals term, $E_{vdW}$ are evaluated, as shown in Figure C.1. We found that both terms show a similar trend as a function of temperature, while the electrostatic term is consistently higher than the vdW term (about 1 to 2 times higher). Thus, the H-bond interaction energy is predominantly electrostatic, as reported in the Charmm forcefield we applied$^{98,136}$. The H-bond interaction energy between silica and C$_6$E$_5$ ($U_{SE}$), and that between C$_6$E$_5$ and H$_2$O ($U_{EH}$) as a function of temperature are illustrated in Figure 6.5c and 6.5d, respectively, as they are responsible for the formation and breakage of the corresponding H-bonds. Further, as we already discovered in section 6.3.1 that the degree of confinement has the most significant effect on the coalescence between the assembly in and out of the pore, we extracted the electrostatic (Ewald term) and van der Waals interaction (LJ term) between the assembly in and out of the pore. We
Figure 6.5. The number of (a) silica-C₆E₅ H-bonds and (b) C₆E₅-H₂O H-bonds as a function of temperature for different $K₁$. The temperature response of H-bond interaction energy (c) between silica and C₆E₅, $U_{SE}$, and (d) between C₆E₅ and water, $U_{EH}$ under different degrees of confinement, $K₁$. (e) Van der Waals interaction between C₆E₅ assembles that are in and out of the pore, $U_{vdw}$, as a function of temperature for different $K₁$. The sudden decrease of $U_{EH}$ together with the increase of $U_{vdw}$ drives the hydrophobic association of the two assemblies in and out of the pore. The lagging response of both $U_{EH}$ and $U_{vdw}$ as a function of temperature leads to the assemblies’ the lower temperature sensitivity under a higher degree of confinement.
found that the electrostatic interaction between the two assemblies is negligible (4% of the LJ term). This observation agrees well with the previous study\textsuperscript{144} that the coalescence of two micelles is a hydrophobic association process between the hydrophobic cores of the two micelles. Thus, we define $U_{vdw}$ (LJ term) as the interaction between those two C$_6$E$_5$ assemblies and plot it as a function of temperature for different $K_1$ in Figure 6.5e. It’s noted that the absolute value of these energetics for different $K_1$ cannot be compared as the number of C$_6$E$_5$ molecules in each simulation box is different. Here we only focus on the change of the $U_{vdw}$ as a function of temperature for each $K_1$.

For $K_1 = 0.23$, 0.5, and 1.0, $U_{EH}$ decrease gradually, followed by a sudden drop near their coalescence temperatures ($T_c = 50$ ℃, 55 ℃, and 60 ℃, respectively), as illustrated in Figure 6.5d. The decrease of $U_{EH}$ initiates the breakage of C$_6$E$_5$-H$_2$O H-bonds, resulting in a similar decreasing pattern of $N_{EH}$ as a function of temperature in Figure 6.5b. The sudden drop of both $U_{EH}$ and $N_{EH}$ at $T_c$ implies that the C$_6$E$_5$ assemblies are quickly dehydrated near this temperature, and they are ready to escape from the H-bond network of the bulk water molecules. (see section 1.2.3)

Meanwhile, $U_{vdw}$ develops a sharp increase at their $T_c$ for all $K_1$ cases in Figure 6.5e. The sudden increase of $U_{vdw}$ initiates the coalescence between the C$_6$E$_5$ assemblies that are located in and out of the pore. Thus, the van der Waals interaction is believed to dominate the hydrophobic association between the non-polar cores of C$_6$E$_5$ assemblies in and out of the pore. This point of view has also been reported elsewhere\textsuperscript{145}, where they show that the van der Waals interaction plays the dominant role in the hydrophobic association of nonpolar ligand and hydrophobic binding sites of the mouse major urinary protein (MUP).

It needs to be noted that in section 1.2.3, it is known that the formation of a single micelle from individual molecules in aqueous bulk solution (without confinement) is due to the entropy-
driven hydrophobic effect. However, here we find that the coalescence (hydrophobic association) of two assemblies is mainly driven by the van der Waals interaction between them. The difference is that in bulk solution without the pore confinement, the surfactant monomers are under the same circumstance. In contrast, in this chapter, the two assemblies are under different conditions, where one of them is under pore confinement, and the other is in bulk solution out of the pore. For the assembly in bulk solution, the number of possible configurations would decrease if it entered into the pore, which will induce the loss of the configurational entropy. The loss of the entropy can be compensated by the attraction from the assembly in the pore. Thus, the migration of the assembly from the bulk space into the pore space and its coalescence with the assembly in the pore is driven by the van der Waals attraction between them. Both this observation and the previously reported work\textsuperscript{145–147} explain the fact that many “hydrophobic associations” in solution do not have to possess the anticipated entropy-driven thermodynamics signature.

Based on the mechanism above, where the decrease of $U_{EH}$ together with the increase of $U_{vdw}$ drive the association of two assemblies, we can further explain the effect of the degree of pore confinement ($K_1$). The change of both $U_{EH}$ and $U_{vdw}$ as a function of temperature at higher $K_1$ is more hysteretic than that at lower $K_1$, leading to a lower temperature-sensitivity of the assemblies under extreme pore confinement (higher $K_1$).

After the coalescence of the two assemblies at $T_c$, the change of H-bond interaction between silica pore and the C$_6$E$_5$ assembly, $U_{SE}$ in the temperature range of $T_c \leq T \leq 80$ °C, directly affects the shape of the assembly. (Figure 6.5c and Figure 6.4a) At $K_1 = 0.23$, $U_{SE}$ first increases then decreases (Figure 6.5c), leading to the shape change of the assembly in Figure 6.4a where the relative shape anisotropy ($K^2$) first increases then decreases. $U_{SE}$ at $K_1 = 0.5$ and 1.0 fluctuates as a function of temperature, as the adsorbed assembly can also feel the attraction from
the opposite pore wall of the narrow pores. This fluctuation results in the variation of the relative shape anisotropy of the assembly adsorbed in narrow pores (Figure 6.4a).

6.6 Summary

In this chapter, we find that the temperature sensitivity of the C₆E₅ assemblies is decided by the temperature response of C₆E₅-water hydrogen bonding (𝑈𝐸Η) as well as the van der Waals interaction between the assembly in and out of the pore (𝑈𝑣𝑑𝑤), and that can be tuned by applying different degrees of confinement, 𝑂₁. The temperature response of both 𝑈𝐸Η and 𝑈𝑣𝑑𝑤 at higher 𝑂₁ is more hysteretic than that at lower 𝑂₁, leading to a lower temperature-sensitivity of the assemblies under extreme pore confinement. In other words, to transport a C₆E₅ assembly from bulk space to the pore space followed by the hydrophobic association of two assemblies, we need to choose a higher temperature for a smaller pore compared to a larger pore. Additionally, the geometric confinement of the cylindrical pore is shown to impact the shape of the adsorbed assembly originating from the silica-C₆E₅ H-bond interaction. The surfactants form a spherical micelle in the larger pore (low 𝑂₁), while they form a rod-like cluster in an ultra-narrow pore whose size is approximately equal to the size of the surfactant monomer. This mechanism makes it possible to design a hydrophobic drug delivery carrier using non-ionic surfactants and program the temperature-responsive delivery process with controllable rates and carrier’s shape by adjusting the pore size.
This chapter summarizes this dissertation's contribution to driving the adsorption and reconfiguration of amphiphilic molecules on silica-based adsorbents. Four projects related to this topic are presented in this dissertation, three of them have been accepted for publication, and the rest is being prepared for submission.

This dissertation demonstrates the role of intermolecular interactions, including electrostatic interaction, van der Waals interaction, and H-bonding in driving the adsorption, reorientation, and morphological transformation of amphiphilic molecules on silica-based adsorbents in aqueous solution. We show that by tuning these intermolecular interactions as a function of external parameters, such as ionization, pH, temperature, and geometric confinement, the amphiphilic molecules, including globular protein, medium-chain fatty acids, and nonionic surfactants, exhibit different adsorbed states. The observations from our simulations agree well with our collaborator’s previous experimental studies, and our results capture the dynamics of molecular binding, self-assembly and reconfiguration of both ionic and nonionic amphiphiles on silica-based adsorbents as well as the molecular driving forces. Thus, this work can not only serve as a suitable proxy in MD simulations to simulate and extract the physics of amphiphilic molecules, but also sets up a basis for choosing the appropriate processing parameters to modulate the structures of amphiphiles for wet bench research.

In Chapter 3, originating from protein-silica surface electrostatic interaction, we aim to control the orientation of cytochrome c after its adsorption. Cytochrome c is a large ionic amphiphilic molecule with an ellipsoid-like native conformation. It is found that the protein-silica electrostatic interaction increases as silica’s degree of ionization is increased, resulting in the increased number of amino acid residues binding to the negatively charged silica surface. The
increased number of binding anchors lead the protein to be adsorbed from a “head-on” to a “side-on” orientation with its native conformation (folded state). Tuning the protein-silica surface interaction not only reorientates the protein but also help to stabilize the protein on a solid substrate while preserving its native conformation. This knowledge provides insights into the immobilization of an enzyme in its preferred orientation for the application of enzyme biosensors, which needs the catalytic site of enzyme to be orientated to the target analyte.

In Chapter 4, we simulate the self-assembly and adsorption process of decanoic acid (FA) at the propylammonium-functionalized silica ($m\text{SiO}_2$)-water interface. The competition between $m\text{SiO}_2$-FA electrostatic interaction ($E_{elec}$) and the FA-FA van der Waals interaction ($E_{vdW}$) governs the ordering/disordering of decanoic acid molecules at the $m\text{SiO}_2$-water interfaces. The balance of these two interactions is found to be corrected to the pH of the decanoic acid solutions, also known as the degree of deprotonation ($R$). At low $R$ ($R < 0.5$ and pH $< pK_a$), the FA-FA van der Waals interaction predominates, driving the decanoic molecules to aggregate to a bulky hydrophobic assembly at the $m\text{SiO}_2$-water interfaces. At higher $R$ ($R \geq 0.5$ and pH $\geq pK_a$), the predominant role of $m\text{SiO}_2$-FA electrostatic interactions over FA-FA van der Waals interactions facilitates the formation of an ordered bilayer adsorbed at the $m\text{SiO}_2$-water interfaces. Based on this knowledge, the external electrostatic attraction is necessary to stabilize a bilayer structure, and that is important in life science research as the fatty acid bilayers are considered as a possible step towards the hypothetical protocells.

In Chapter 5, we study the temperature-responsive adsorption behavior of ethoxylated surfactants, $C_6\text{E}_3$ and $C_6\text{E}_5$ in a cylindrical pore of diameter 8.6 nm in the range of $20 \, ^\circ\text{C} \leq T \leq 45 \, ^\circ\text{C}$. We find that the adsorption of the both surfactants are initiated by the formation of silica-surfactant H-bonds. Both silica-$C_6\text{E}_3$ and silica-$C_6\text{E}_5$ H-bond interactions are nearly independent
of temperature, enabling part of the strongly adsorbed surfactants to serve as the nucleation sites on the pore wall. The decrease of surfactant-water H-bond interaction as a function of temperature induces the breakage of surfactant-water H-bonds, driving the condensation of surfactants at the nucleation sites on the pore wall. While both C₆E₃ and C₆E₅ molecules show temperature-responsive aggregative adsorption at the pore wall, they demonstrate different temperature sensitivity due to the difference in their hydrophilicity (number of oxyethylene groups). The weaker C₆E₃-water H-bonding has a more instantaneous response to temperature, leading to a higher temperature sensitivity of C₆E₃ assemblies compared to C₆E₅ assemblies. Thus, parameters that influence the silica-CₙEₘ and CₙEₘ-water H-bonding can be used to program CₙEₘ’s adsorption behavior and thermal stability. For example, with increasing temperature, CₙEₘ with a higher m value features a slow aggregative adsorption process with higher thermal stability. In comparison, CₙEₘ with a lower m value is more thermal sensitive and less thermal stable. Additionally, a competitive molecule can be introduced to interrupt the silica-CₙEₘ H-bonding to hinder the adsorption or induce the desorption of the CₙEₘ molecules. It also makes the design of complex structures possible by judiciously choosing the CₙEₘ molecules with different hydrophilicity.

Further, in Chapter 6, we continue studying temperature-induced adsorption and aggregation of C₆E₅ in cylindrical pores with the emphasis on the effect of pore size. The decrease of pore size is a direct indicator of the increase of the geometric pore confinement. We find that the varying pore size mainly affects the later stage of the aggregation process, where the assembly outside of the pore migrates into the pore to coalesce with the one in the pore at a higher temperature. The decrease of C₆E₅-water H-bonding (𝑈_{E𝐻}) together with the increase of van der Waals interaction between the assembly in and out of the pore (𝑈_{v𝑑𝑤}) as a function of temperature
drives the hydrophobic association of these two assemblies. The association in silica pores of varying size occurs at different temperatures, and the temperature upon the completion of the coalescence is defined as the coalescence temperature, $T_c$. The thermal responses of $U_{EH}$ and $U_{vdw}$ are more hysteretic as the pore size is decreased, leading to a higher $T_c$ value in a narrower pore. Based on this knowledge, we can program the temperature-responsive transportation of the ethoxylated surfactant assemblies by tuning the pore size. The thermo-responsive transportation and coalescence of ethoxylated surfactants in porous silica materials allow the encapsulation and release of hydrophobic drugs with appropriate heating/cooling programming. Given that $T_c$ increases with the decreasing pore size, we predict that the drug delivery rate will decrease simultaneously, which is of great significance in developing new carrier for the drug delivery with a controlled rate.
APPENDIX A. SUPPLEMENTAL FIGURES FOR CHAPTER FOUR

Figure A.1. Probability distribution of the alkyl chain length of fatty acid molecules (a) in bulk water and (b) adsorbed on $m$SiO$_2$ as a function of $R$.

Figure A.2. The density profiles of (a) all atoms and (b) headgroups (oxygen in $-\text{COOH}$ and $-\text{COO}^-$ groups) in the characteristic assembly. (c) Distribution of the angle between fatty acid molecule ($C_{10}$-$C_1$ vector) in the characteristic assembly and $m$SiO$_2$ surface normal.
Figure B.1. Electrostatic contribution, $E_{elec}$ and van der Waals contribution, $E_{vdW}$ to (a) silica-water H-bonding, (b)(c) silica-C$_6$E$_m$ H-bonding and (d)(e) C$_6$E$_m$-water H-bonding as a function of temperature.
Figure C.1. Electrostatic contribution, $E_{\text{elec}}$ and van der Waals contribution, $E_{\text{vdW}}$ to (a-c) silica-C$_6$E$_5$ H-bonding (d-f) C$_6$E$_5$-H$_2$O H-bonding as a function of temperature under different degrees of confinement, $K_1$. 
APPENDIX D. LETTERS OF PERMISSION

Figure 1.1 is adopted from Ref.23 with the permission of Elsevier, as shown below.
Figure 1.4a and 1.4b are reproduced from Ref.\textsuperscript{41} with the permission of Elsevier.
Figure 5.2a-d, and Figure 5.4a-c are reproduced from our previous publication titled “Directed Pore Uptake and Phase Separation of Surfactant Solutions under Confinement” with the permission from American Chemical Society, as shown below.
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