Molecular Dynamics Modeling and Simulation of Chromatographic Bioseparation

Jee-Ching Wang and Athanasios I. Liapis, University of Missouri-Rolla, Rolla, MO

Recent advances in molecular biology and separation science have resulted in large-scale production of diagnostics and biotherapeutics. In downstream purification of the produced biomacromolecules, desirable bioseparation performance can be achieved using chromatography processes based on suitable supported matrix and affinity ligands. Because the biomacromolecules and the affinity ligands possess partial charge distributions and can even be charged, the mass transport of a biologically active charged macromolecule (i.e., peptide or protein) in an electrolytic solution in contact with a charged surface onto which the charged macromolecule is adsorbed, represents a process of very significant importance in chromatographic separations as well as in the functioning of biological systems. In addition, such transport and interaction mechanisms have to be considered in food processing, bacterial cell adhesion and in the development of biomaterials and biosensors as well. However, many important aspects of these mechanisms have not been properly considered in many existing models, where the electrically driven mechanisms are either ignored or lumped into other phenomenological terms, making the models unsuitable for explaining certain new but important phenomena such as concentration overshoot in the adsorption of charged molecules in porous adsorbent particles.

Experimentally the kinetic and equilibrium characteristics of the mass transport and adsorption of charged solutes onto different types of charged solid surfaces have been traditionally studied by analyzing uptake and breakthrough curves obtained from finite bath (batch) and column experiments, respectively. However, these methods only provide an indirect measurement of the concentration and transport rate of the charged adsorbate within the charged adsorbent particles. In order to properly describe and predict the dynamic behavior of the chromatographic bioseparation systems for the time and length (size) scales employed in practice, it is of paramount importance that the constructed macroscopic continuum models properly consider the major in contributing significance transport and interaction mechanisms involved in such systems and employ constitutive expressions that appropriately describe the behavior of these mechanisms. The field of molecular dynamics simulation provides a microscopic modeling approach that could be used to determine the kind and relative importance of the mechanisms involved in the transport of a biologically active charged macromolecule in an electrolytic solution and its interaction with an affinity ligand and a substrate surface. To this end, molecular dynamics simulation studies are conducted with atomistic potential models in this work to determine the mass transport of model charged biomolecules and their adsorption onto a charged surface.

Our simulation system is an interfacial system comprising a charged solid surface in contact with a liquid solution that contains a peptide molecule (desmopressin), water molecules, cations, and anions. The model biomolecules is desmopressin, which consists of nine amino acid residues. For computational accuracy and efficiency, we use both full-atomic and united atom models to simulate desmopressin. Specifically, the -CH3-, -CH2-, and -CH- groups are modeled as united atoms (UA's) with interaction centers placed at the positions of carbon atoms, while the rest H, N, O, S, and C atoms are modeled as individual atomistic interaction sites. All these atoms carry partial charges and the net charge of desmopressin is

+1e. Each bond in the desmopressin molecule is constrained to its equilibrium length by applying the SHAKE algorithm. To retain the realistic molecular structure, other intra-molecular interactions are needed, which include bond angle bending, dihedral torsion, and non-bonded van der Waals (vdW) and electrostatic interactions. In this study, the aqueous electrolyte solution is represented by 4494 water molecules that are explicitly simulated using the TIP5P model. This newly developed model treats a water molecule with five interaction sites, including a charge neutral oxygen atom, two hydrogen atoms, and two electron lone pairs. Each H atom carries +0.241e charge and each electron lone pair is modeled as a point charge carrying -0.241e charge. Compared to several other popular models, the TIP5P model gives a better representation of water densities and of the water structure under ambient conditions. As in typical peptide and protein systems, the cation and anion considered in this study are sodium ion (Na+) and formate ion (HCOO–). Nine sodium ions and nine formate ions are simulated and modeled as charged spherical particles. The corresponding ionic concentration is 100 mM. The model solid surface is taken to be a negatively charged flat surface whose lateral dimensions are 53.04 Å×53.04 Å and periodic boundary conditions are applied along the x and y directions. The surface has a fixed charge density of –0.0228 C/m^2, which is represented by four –e charges uniformly distributed on the model surface.

The molecular dynamics simulations of this work have shown that the structure of the charged macromolecule (desmopressin) changes as it approaches the charged solid surface and the effect of the charged solid surface on the structure and transport coefficient of the charged macromolecule increases significantly as the charged macromolecule enters the electrical double layer adjacent to the charged solid surface. The significant increase in the value of the transport coefficient of the charged macromolecule above the value of its diffusion coefficient in the bulk liquid phase as the macromolecule approaches the charged solid surface, was found to be due to the strong electrostatic interaction between the charged macromolecule and the charged solid surface; this finding strongly supports the very important role that the mechanism of electrophoretic migration plays in the transport of a charged solute in the region of the electrical double layer adjacent to the charged solid surface. In addition, it was found that explicit solvent has to be employed otherwise the magnitude of the potential energy of the interaction between the adsorbed charged macromolecule and the charged solid surface, as well as the structure of the adsorbed macromolecule lack real physical representation. The dominant potential energy involved in the adsorption of the charged macromolecule onto the charged solid surface is the electrostatic (Coulombic) potential. The large magnitude of the electrostatic interaction potential together with a negligible probability for the possibility of many water molecules in the surface hydration layer undertaking concerted moves cause the values of the transport coefficients of the center of mass of the adsorbed macromolecule along the lateral *x* and *y* directions on the charged solid surface to be about equal to zero.

We also studied the conformations and the values of the lateral transport coefficient of desmopressin in the adsorbed layer and in the two liquid layers located above the adsorbed layer with molecular dynamics simulation. The results show that while the lateral mobility of the adsorbed desmopressin is approximately equal to zero, the lateral transport coefficient of the biomolecule in the liquid layers above the adsorbed layer increases as the distance of the location of the liquid layer from the charged solid surface increases. Furthermore, the values of the lateral transport coefficient of the biomolecule in the liquid layers above the adsorbed layer are lower than the value of the transport coefficient of desmopressin along the z direction

(which is normal to the charged solid surface) in the liquid phase located above the vacant charged sites of the solid surface. These differences in the transport coefficients imply that the contributions of diffusion and electrophoretic migration on the transport of desmopressin in the liquid layers parallel to the adsorbed layer and in the liquid phase above the vacant charged sites and in the direction normal to the solid surface, are different and have important implications with respect to the replenishment of the desmopressin molecules in the inner parts of a channel (pore) and overall rate of adsorption. More specifically, a lower bound of the linear characteristic dimension (i.e., pore radius) of a pore (channel) exists for efficient replenishment and separation of biomolecules. We demonstrated that this lower bound could be obtained from relevant information provided by simulation studies and this estimate is critical for the size of the radius that the pores of a porous adsorbent particle or of a skeleton of a porous monolith should have in order to realize efficient separation with respect to transport and overall adsorption rate. The estimation of the value of the lower bound of the linear characteristic dimension of a pore in porous adsorbent particles or porous skeletons of a monolith through the information determined from the molecular dynamics simulation (microscopic modeling approach) provides necessary and useful information about the appropriate size of pores that have to be considered in investigations that employ macroscopic models for studying the behavior of the adsorption of a biomolecule in adsorbent media as well as in the construction of such media.