JST Basic Research Programs

C R E S T

(Core Research for Evolutional Science and Technology)

Annual Report for Research Work in the fiscal year 2006

Research Area :

High Performance Computing for Multi-scale and Multi-physics Phenomena

Research Theme

Research and Development of DDS Simulator

Name of Research Director, Belonging and Title:

Masuhiro Mikami, Research Institute for Computational Sciences(RICS), National Institute of Advanced Industrial Science and Technology(AIST), Deputy Director

§1. Outline of Research Work

The drug transportation only to the body part affected by illness is very important to increase the drug efficacy and to reduce adverse drug reactions. Therefore, development of a carrier which incorporates drugs is of high priority and such drug delivery system (DDS) presents a multi-scale and multi-physics problem which consists of the three research topics. (a) The design of the DDS nanoparticles (the complex of the liposome and a sugar chain) which incorporate a drug molecule, (b) the molecular design of the sugar chain which recognizes lectins in the vessel wall near the diseased part, (c) the design of the transport process of the DDS nanoparticles in the blood vessel. The design technology of DDS is not yet established and its development proceeds in the groping fashion. In this research project, we will develop a multi-scale simulation methodology for the active targeting drug delivery system consisting of a liposome and a sugar chain. Also, we will provide the basis for a general DDS design by developing a DDS simulator which integrates these simulation methodologies.

§2. Content of Research Work

In this research project, we will develop a multi-scale simulation methodology for the active targeting drug delivery system (DDS) consisting of a liposome and a sugar chain. This simulation methodology based on the fragment molecular orbital method, molecular simulation and fluid dynamics enables us to do (1) design of DDS nanoparticle, (2) analysis of molecular interaction between lectin protein and sugar chain, and (3)flow analysis of DDS nanoparticles in a blood vessel, which is essential for the DDS development shown in Fig.1. In this current year, we employed postdoctoral fellows to strengthen our research group and purchased the parallel computer system for the actual research work starting 2007. Also, in order to obtain the most effective and fruitful results on the present five-year project starting from October 2006, we have (1) made a global survey on the work related to DDS, and (2) examined the existing methods and the available numerical simulators.

(1)Research and development of molecular simulation for design of DDS nanoparticle(G1)

The information about the size and stability of the DDS nanoparticle and the unit density of the sugar chain attached to the surface is necessary to develop the DDS nanoparticle, which transports a drug only to the affected part. Therefore, we will develop the molecular simulation methodology enabling us to investigate the factors (surface tension, elasticity, temperature and the concentration of the lipid molecule etc.) which control the size and stability of the DDS nanoparticles. In this current year, we carried out the following surveillance studies, and decided upon the strategy of the actual research starting 2007. (1) We have investigated the published papers reporting the basis set and electron correlation effects on calculated interaction energies of molecules related to lipid-lipid and lipid-water interactions. We found that a large basis set and the MP2 level electron correlation

correction are necessary for studying the intermolecular interactions between lipid molecules. (2) DDS nanoparticles are composed by glycerophospholipid, cholesterol, and sphingoglycolipid to increase the stability. In such a membrane, a phase separation occurs with the increase of the concentration of cholesterol, and domains are formed. According to the recent experimental work, the domain influences the stability of DDS nanoparticles, however, the structure and properties are still unknown. Therefore, our research target is to investigate the structure and properties of the membrane, and we have already optimized the potential parameters and initial coordinates, and undertook practical simulations. (3) In order to investigate the dynamical process of the liposome formation and the relationship between the various physical properties and the structure of lipid molecules, we have tried to develop coarse-grained model systems and analysis programs. Lipid molecules are represented by the combination of hydrophilic beads and hydrophobic beads with various molecular geometries. Since the solutions of these kinds of amphiphilic molecules such as micelles, membranes and vesicles show complicated phase diagrams, we first searched the optimum simulation conditions that lead to the liposome formation.

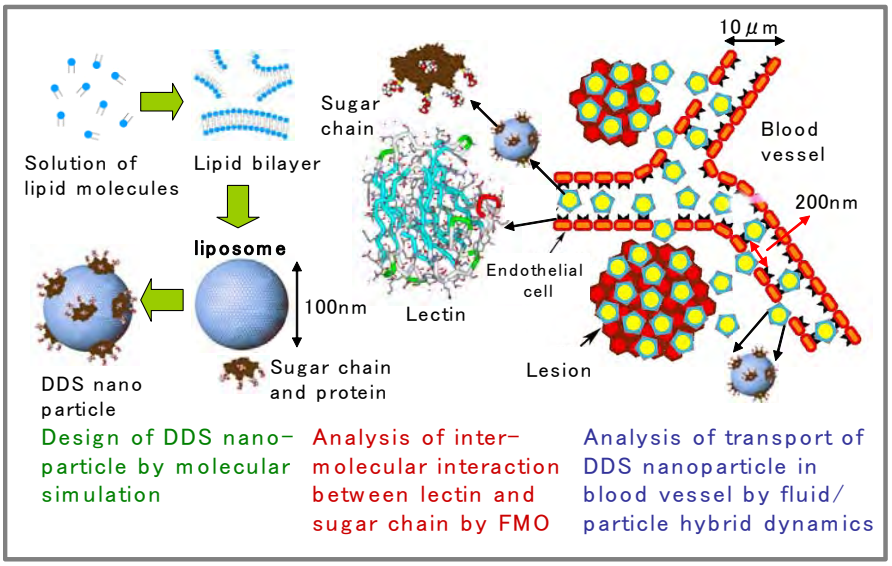

Fig. 1 Conceptual diagram of the DDS simulator

(2)Research and development of analysis method of molecular interaction between lectin protein and sugar chain(G2)

Carbohydrate-protein interactions are ubiquitous in cell-cell recognition process. In contrast to the rapid progress of glycobiology, in many cases, the molecular basis of carbohydrate recognition by target lectins remains elusive mainly because of the lack of high-resolution structural data for the complexes. Among many lectin families, the selectins are responsible for the early adhesion events of the leukocytes onto the endothelial cells. To understand the molecular origin of the tethering and rolling process by leukocyte, we analyze the binding process of selectin with sialyl lewis x (sLex)

epitopes based on the systematic computational modeling. The strategy of our calculations is composed of three part: (1) MD simulations for the selectin-sLex complex are conducted to sample the bioactive conformations in the solution phase, and then (2) ab initio QM/MM structural refinements are performed to clarify the detailed information of the binding epitope, (3) finally we evaluate the molecular interaction energy based on the all electron quantum chemical calculations for the entire complexes, by using the fragment molecular orbital (FMO) method.

At this period, we have performed the structural optimization of E-selectin-sLex complex with QM/MM RHF/6-31G*/AMBER calculations. Although the sLex epitope binds weakly onto the surface of the c-type lectin domain, we cannot observe the large structural deviation from the original X-ray data. By performing the interaction energy analyses based on the QM/MM as well as FMO, we determined the amino acid residues crucial for molecular recognition. The residues that have high affinity for sLex are limited to several Glu and Arg located on the binding region, and both QM/MM and FMO predict qualitatively similar results. This result clearly implies that the essence of carbohydrate-protein interactions is of the electrostatic nature.

(3)Research and development of flow analysis of DDS nanoparticles in a blood vessel (G3)

We have firstly reviewed a so-called drug delivery system developed by other research works, and elucidated the present status and the problems to be solved. Secondly we have surveyed the fluid flow simulators available to our research project; the latter aims at transporting nano-scale liposomes containing drugs by means of blood flow. We found that some of the numerical simulators can be used for the analysis and display of our blood flow at the macroscopic level. Thirdly, we have collected clinical data on the formation of a tumor, as well as associated interstitial flow changes. Lastly we have accumulated evidence on anatomical structure of the luminal surface of the vascular endothelium. We found that the presence of the layer, called glycocalyx, can have a profound effect on the filtration of liposomes. In order to enhance the "effectiveness" in our DDS, to have the closest joint research links with G1 and G2 groups, and to show the advantage of our research over other similar projects, we arrive at the strategy that we should concentrate on (i) the processes in which the liposomes approach and permeate the luminal surface of the vascular endothelium, taking into account the work by our Group 2, and (ii) the processes in which a new channel of the blood flow into a tumor cell is created.

At present, we have already calculated a network formation of the blood flow around a tumor cell, considering the nonlinear process of the flow concentration because of the presence of such self-reproducing cells and the resulting changes of the boundary shape due to the tissue destruction. The possibility of the enhanced liposome transportation to a target cell has been presented at domestic symposia. We are also preparing the computational environment, involving the newest computing system available at AIST (National Institute of Advanced Industrial Science and Technology), which enables us to develop our research project. A test of available software to

analyze the blood flow and to calculate the interaction between glycocalyx and liposomes is in progress.

§3. Formation of Research Work

(1)AIST group

	Name	Belonging	Title
○	Masuhiro Mikami	RICS*, AIST**	Deputy Director
○	Kazuo Kitaura	RICS*, AIST**	Principal Research Scientists
	Tadafumi Uchimaru	RICS*, AIST**	Group Leader
	Seiji Tsuzuki	RICS*, AIST**	Senior Research Scientists
	Yuto Komeiji	RICS*, AIST**	Senior Research Scientists
	Toshiaki Miura	RICS*, AIST**	Senior Research Scientists
	Dmitri Fedorov	RICS*, AIST**	Research Scientists
	Tetsuya Morishita	RICS*, AIST**	Research Scientists
	Toyokazu Ishida	RICS*, AIST**	Research Scientists
	Kengo Nishio	RICS*, AIST**	Research Scientists

* Research Institute for Computational Sciences

** National Institute of Advanced Industrial Science and Technology

Items of Research

(1)Research and development of molecular simulation for design of the DDS nanoparticle

(2)Research and development of the analysis method of molecular interaction between lectin protein and sugar chain

(2)TUAT group

	Name	Belonging	Title
○	Osamu Sano	Institute of Symbiotic Science and Technology, Tokyo University of Agriculture and Technology	Professor
*	Yusaku Nagata	Institute of Symbiotic Science and Technology, Tokyo University of Agriculture and Technology	Post-doctoral researcher

Items of Research

Research and development of flow analysis method of DDS nanoparticles in a blood vessel

(3)TOSHIBA group

	Name	Belonging	Title
○	Satoshi Itoh	Corporate Research & Development Center, TOSHIBA CORPORATION	Chief Research Scientist

Items of Research

Research and development of flow analysis method of DDS nanoparticles in a blood vessel

§4. Publication of Research Results

（4－1） Publication of Thesis (The original Work)

① Number of Publications (0 times-Domestic, 0 times-International)

② Detailed Information of Thesis

（4－2） Patent Application

① Cumulative Number

1) Patent Applications in the fiscal year 2006 (Domestic- 0 Cases, Oversea- 0 Cases)

2) Cumulative number of Patent Applications for the research period of CREST
(Domestic- 0 Cases, Oversea- 0 Cases)