Accelerating the Research in Drug Delivery System; A Challenge of the Earth Simulator to Medical Innovation

Project Representative
Atsushi Miyauchi Research Organization for Information Science and Technology

Authors
Atsushi Miyauchi *1, Shogo Tejima *1, Aya Marumo *1, Hisashi Nakamura *1, Yuichi Yamasaki *2 and Kazunori Kataoka *2

*1 Research Organization for Information Science and Technology
*2 University of Tokyo

Objectivity of our project is to help the research of drug delivery system by means of modern high performance computers. Up to the last year we have carried out some large-scale simulations on the system of DNA and PEG-PLL. On the other hand, a polymer family of PEG-PAsp (DET) is getting to be thought of as promising candidate for in-vivo application recently. However, it still needs improvements in condensation. So this year we performed some simulations on PEG-PAsp (DET) in solution. In those calculations, package programs of classical molecular dynamics was employed to take a large number of molecules into account. The results showed that adding cholesterol helped polymers to get together faster and make stable cluster.

Keywords: drug delivery system, molecular dynamics, micelle, poly-aspartic acid, poly-ethylene-glycol

1. Introduction

Since the end of the last decade nanotechnology has been grown at rapid pace and spread over many different fields out of its birthplace, material science. Now in medical science, nanotechnology is expected to open up the door to the innovative methods of treatments no one can imagine twenty years ago. One of its novel application is drug delivery system (DDS). The main purpose of DDS is to target the seat of a disease and carry drugs there precisely.

Our project aims to accelerate the research of DDS by fully exploiting the amazing computational ability of the Earth Simulator. Up to the last year we have carried out some large-scale simulations on the system of DNA and block copolymers comprised of poly-ethylene-glycol (PEG) and poly-L-lysin (PLL). On the other hand, a polymer family of PAsp (DET) is getting recognized as promising candidate for practical application for its relatively gentle damage to human body [1]. However, it still needs improvements especially in condensation. Recently Oba et. al. reported that adding cholesterol as a hydrophobic base enabled PAsp(DET) to enhance condensation in water significantly [2]. To reproduce their experimental results numerically we performed some simulations. Since we thought classical molecular dynamics was suitable to take a large number of molecules into account, we used the program package Amber for its reliability [3]. The results showed that adding cholesterol seemed to help polymers to aggregate and to make stable cluster.

This report is organized as followings. The next section describes the physical aspects of DDS in some detail. In the third section, we discuss computational methods. The forth section gives computational results. Final section summarizes this report.

2. Drug delivery system

Many kind of DDS technology has been proposed so far. Amongst them, our project pays much attention to the method using nano-sized particle called micelle mainly comprised of PEG-PAsp (DET) block copolymers. This method was recently developed by Professor Kataoka of the University of Tokyo and is expected to be promising in near future gene delivery because of its relatively low impact on human body [1]. Although the whole process of PEG-motivated DDS is much complicated, we can recognize four characteristic stages in that. See Fig. 1.

In the first stage, relaxed DNA attached to a PEG-PAsp (DET) copolymer starts to condense in solution. Shortly after, DNA is condensed to a small object like ball. Notice that PEG-PAsp (DET)-DNA complex has a hydrophilic end in PEG and a hydrophobic end in DNA. In short, it has amphipathic property. In the next stage, hundreds of those PEG-PAsp (DET)-DNA complexes in the water meet

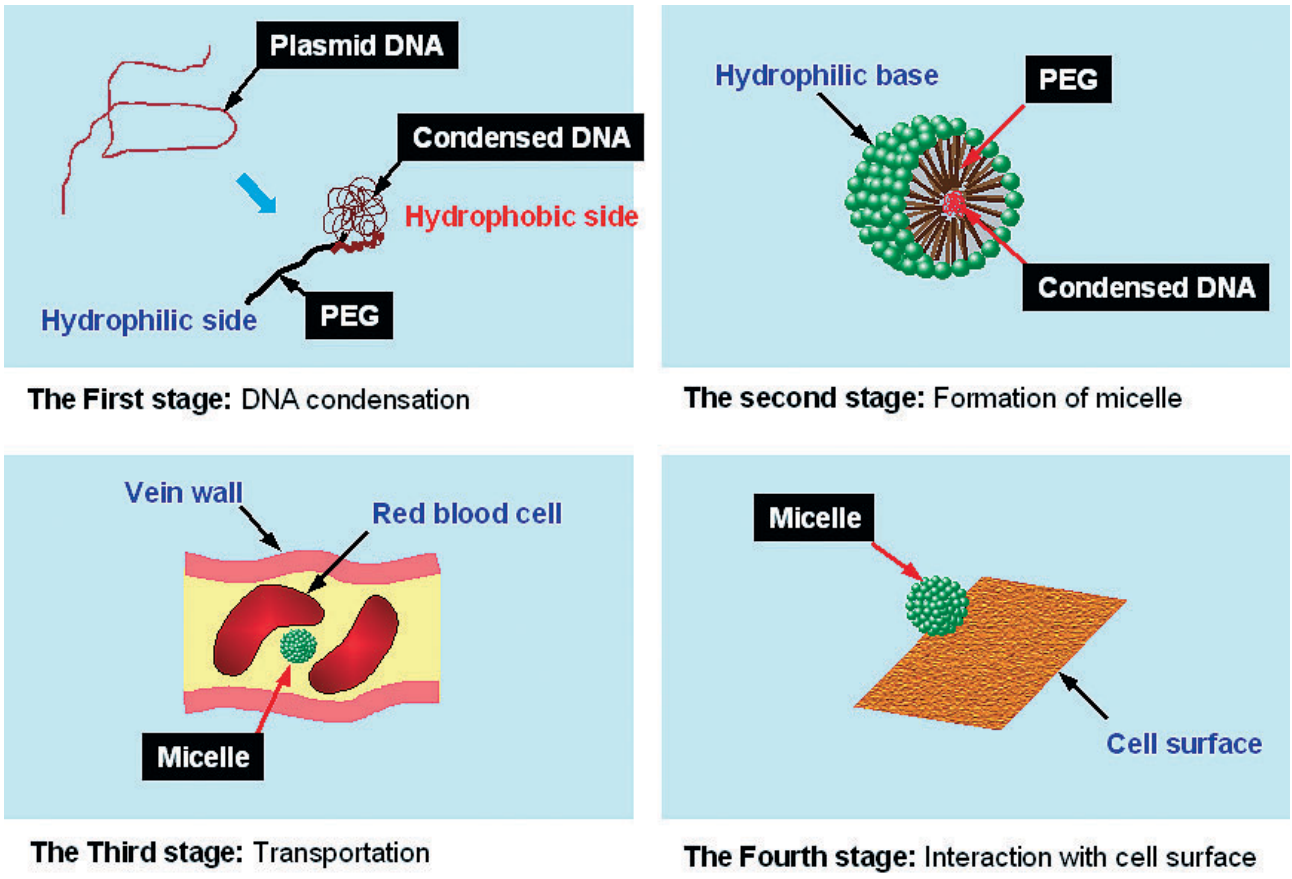

Fig. 1 Characteristic stages of drug delivery system.

together and spontaneously form sphere called micelle in which each condensed DNA heads for the center and PEG heads for the surface. The driving force of this self-organizing formation is amphipathic property mentioned above. In the third stage, micelles are carried through vein and capillary tube slipping through the red blood cells. In the last stage, micelles reached the targeted portion of disease are attracted by the local gradient of ion concentration in the vicinity of the cell surface and then absorbed into it through the carrier or channel protein located at the membrane.

Those types of problem are recently paid attention in many fields and classified as multi-scale and multi-principle phenomena in contrast to single-scale and single-principle phenomena conventional science has been dealing with. To simulate such kind of complicated phenomena efficiently we must choose effective computational methods corresponding to each stage and combine them interactively. However, in this report, we should focus our interest on the second stage, i.e. micelle formation. That kind of problems can be treated using well established technique such as classical molecular dynamics.

3. Computational methods

Besides tens to hundreds of polymer molecules, thousands to millions of water molecules participate in micelle formation. Tracking the motion of such large number of molecules quantum mechanically is not easy task even for the latest high-end computer systems. Among the current simulation techniques, solely classical molecular dynamics can be served as workhorse for such problems. In this paper, we used Amber developed by UCSF, which is known as the most widely used and well verified program package [3]. The chemical formulas of PEG-PAsp (DET) and PEG-PAsp (DET)-Chole are shown in Fig. 2 and Fig. 3. At first, these formulas should be described in computer readable format. We used ChemSketch for this purpose [4]. Its output files were in .mol format. For the convenience of following process, those files were converted in .pdb format through Converter [5]. Resultant .pdb files were fed into Antechamber, one of utility programs of Amber package, to make force field files in .prep format. Then .prep files were fed into Leap, another utility of Amber package, to replicate molecules and to add waters. We prepared two simulation cases, one for PEG-PAsp (DET) and another for PEG-PAsp (DET)-Chole. Each employed eight polymers, two in a row, four in a column. Distance between rows was 20 angstrom, columns 30 angstrom, respectively. The least distance from molecules to boundary was set 12 angstrom. In addition, standard TIP3 water was used. The dimensions of resultant bounding box and total number of atoms were 80 × 95 × 95 angstrom and 66,118 atoms for PEG-PAsp (DET) and 80 × 105 × 105 angstrom and 78,089 atoms for PEG-PAsp (DET)-Chole. Consequently, two sets of input files for MD calculation were generated, i.e. .top and .crd. Before MD cal-

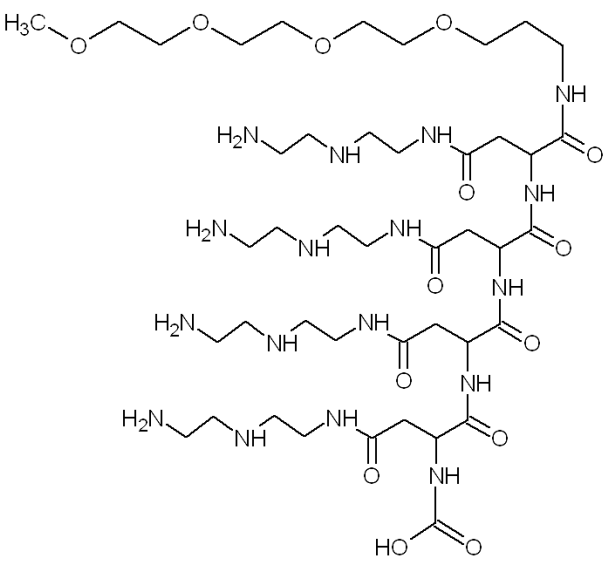

Fig. 2 Chemical formula of PEG-PAsp (DET).

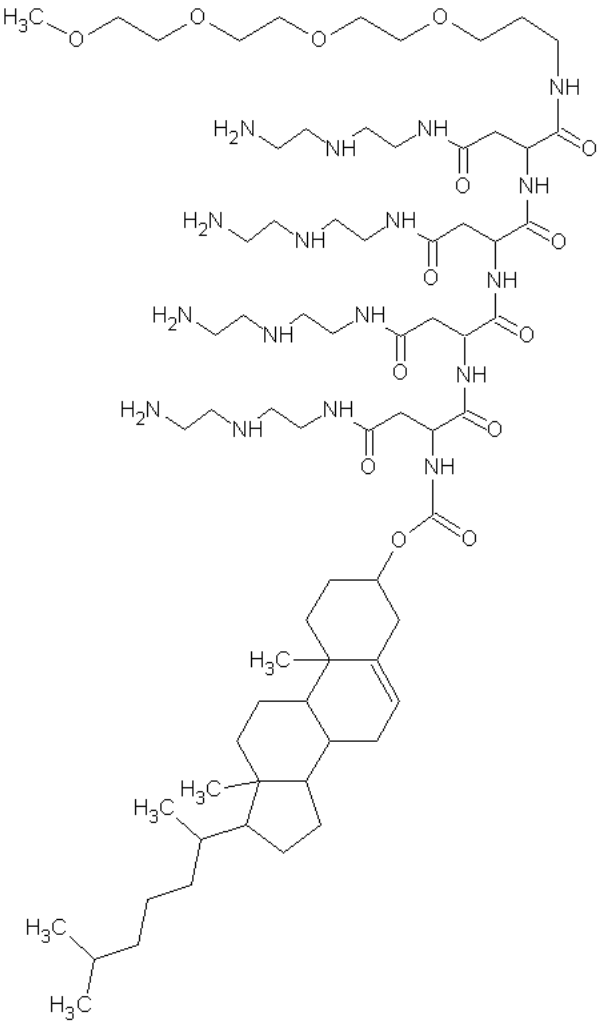

Fig. 3 Chemical formula of PEG-PAsp (DET)-Chole.

culations, we performed 500 energy minimization steps to get rid of initial inconsistency and to set temperature at 300 K. After those preparations, we could start MD calculations at last. Temporal increment was set as 1 fs, which is recommended for standard use. Typical time for bio-molecules is thought to be micro to mille-seconds. It requires billion to trillion iterations and is far beyond the capabilities of the current fastest supercomputer. So making a compromise with available resources, we determined to calculate one million iterations, i.e. 1 nano-second. Since job running at a time was restricted in time. So we restarted job twenty times. Calculated data were reconverted in .pdb format using Amb2pdb, one of Amber utility programs. Finally .pdb formatted files were visualized using Jmol viewer [6].

4. Results

We carried out two long simulation runs as mentioned above. Results of each run are described in the following subsections.

4.1 PEG-PAsp (DET)

Figure 2 depicts the chemical formula of PEG-PAsp (DET). In this figure, we can see PEG consists of four ethylene molecules on the top-side, four Diethylenetriamine (DET) chains below it and a backbone on the right-hand side which consists of four Aspartic acid bases. Figure 4 shows the initial configuration of computational domain. The entire

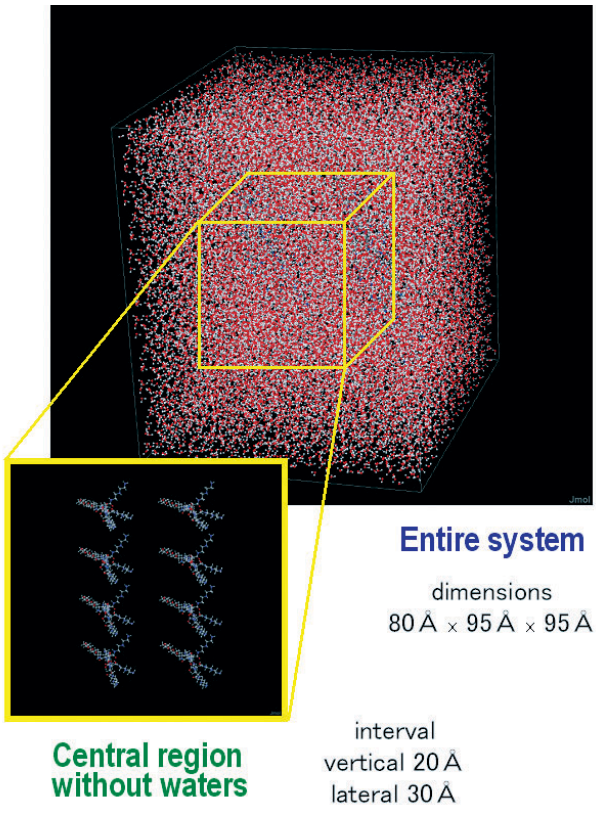

Fig. 4 Initial configuration of PEG-PAsp (DET).

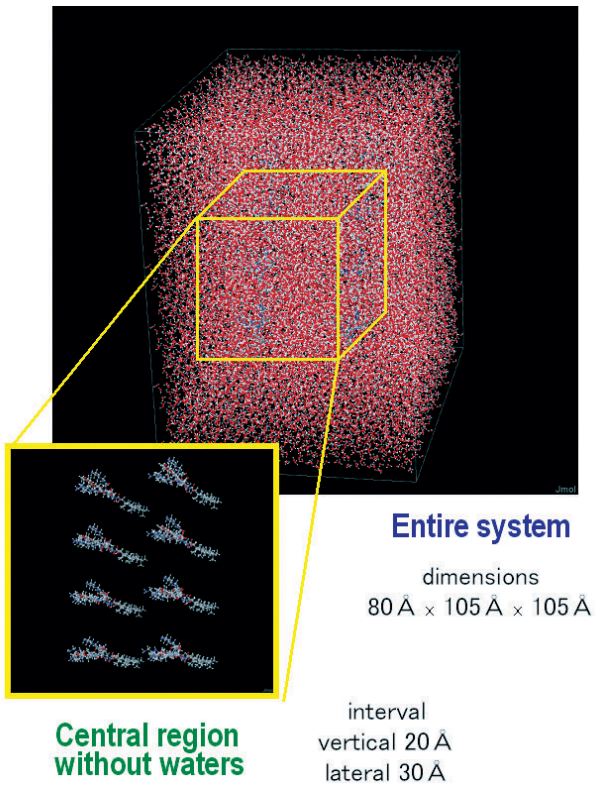

Fig. 5 Results of PEG-PAsp (DET) at t = 1.0 ns.

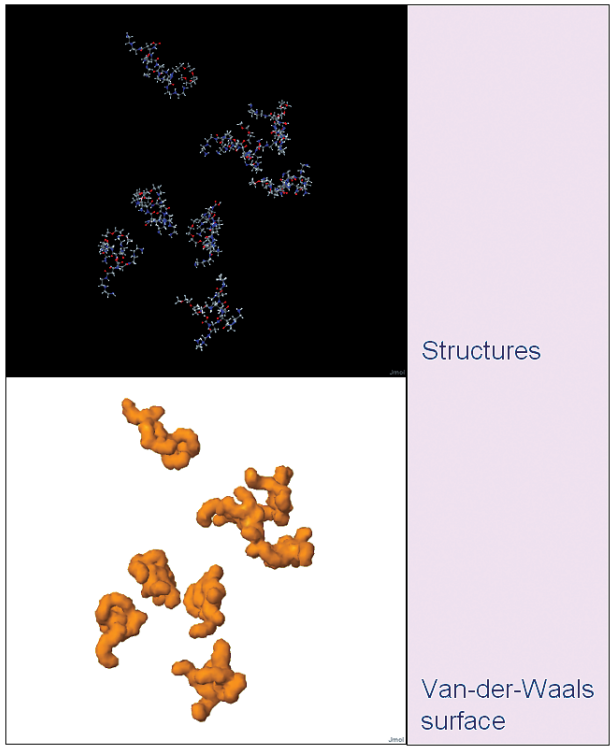

Fig. 6 Initial configuration of PEG-PAsp (DET)-Chole.

system contains eight polymer molecules and 21,610 water molecules, i.e. 66,118 atoms. This initial configuration was obtained after 500 energy minimization steps. Figure 5 shows the results after 1 ns. Water molecules are hidden to see polymer molecules clearly. From Van-der-Waals surface, we can see three molecules seem to be loosely connected in upper-right while the rest five molecules remains isolated. However, close inspection of structure reveals that aggregated three molecules are just touching toe each other.

4.2 PEG-PAsp(DET)-Chole

Figure 3 depicts the chemical formula of PEG-PAsp(DET)-Chole. In this figure, we can see PEG, four DET chains and Aspartic acid bases just the same way above. In addition, here exists a large cholesterol molecule at the end of backbone. Remind that PEG is hydrophilic while cholesterol is hydrophobic. Consequently PEG-PAsp(DET)-Chole has strongly amphipathic property. Figure 6 shows the initial configuration of computational domain. The entire system contains eight polymer molecules and 25,651 water molecules, i.e. 78,809 atoms. This initial configuration was obtained after 500 energy minimization steps. Figure 7 shows the results after 1 ns. Water molecules are hidden to see polymer molecules clearly. From Van-der-Waals surface, lower five molecules seem to aggregate like single molecule. In fact, from structures, we can see right three molecules are tightly tangling with their gray-colored cholesterol parts, as well as left two molecules.

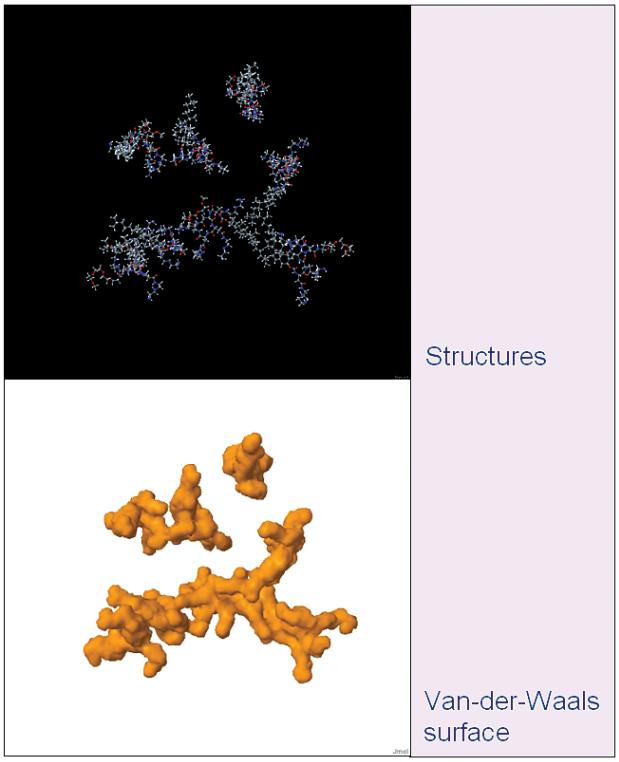

Fig. 7 Results of PEG-PAsp (DET)-Chole at t = 1.0 ns.

5. Conclusion

Comparing the results of PEG-PAsp (DET) with PEG-PAsp (DET)-Chole in previous section, we can conclude that the addition of cholesterol enables significant improvement in condensation. Our conclusion approves Oba's interpreta-

tion of his experiments to a certain extent. It is important fact that simulation can give us insight in some extent in DDS research. However, notice that our simulations are rather small compared to practical micelles. Integration time is also shorter than practical phenomena. Furthermore, system dimensions should be larger to suppress the periodic boundary effects arise from particle mesh Ewald method. Computational resources to fill those requirements are probably beyond current TFLOPS systems. Further sophistication might be made on next generation PFLOPS systems.

References

[1] K. Kataoka, "Developments of polymer-micelle type nano-devices toward realization of gene therapy", J. Pharmaceutical Science and Technology Japan, Vol.68 supplement, p50, May 2008 (in Japanese).

[2] M. Oba et. al., "Developments of polymer-micelle type gene-vectors utilizing hydrophobic interaction", J. Japan Society of Drug Delivery System, Vol.23, No.3, p417, May 2008 (in Japanese).

[3] http://ambermd.org/

[4] http://www.acdlabs.com/products/chem_dsn_lab/chemsketch/

[5] http://www.molecular-networks.com/software/convert/index.html

[6] http://jmol.sourceforge.net/

地球シミュレータを利用したドラッグデリバリシステムの研究：革新的医療への挑戦

プロジェクト責任者
宮内　敦　高度情報科学技術研究機構

著者
宮内　敦*1，手島　正吾*1，丸茂　彩*1，中村　壽*1，山崎　裕一*2，片岡　正則*2
*1　高度情報科学技術研究機構
*2　東京大学

本プロジェクトの目的は高性能計算機を用いることでドラッグデリバリシステムの研究を加速することにある。昨年度まではDNAとPEG-PLLからなる系の大規模シミュレーションを実施した。しかし最近、PEG-PAsp (DET) が生体応用への有力な候補と見做されるようになってきた。半面、凝縮能にまだ改良の余地があるともされる。そこで本年度は水溶液中のPEG-PAsp (DET) の振る舞いをシミュレートした。計算には多数の原子を考慮するため、古典分子動力学のパッケージであるAmberを用いた。その結果、コレステロールを付加することでミセルへの凝縮がより速く安定的であるという結果が得られた。

キーワード：ドラッグデリバリシステム, 分子動力学, ミセル, ポリアスパラギン酸, ポリエチレングリコール