Collaboration Projects

Molecular Transport System in Molecular Communication

We have successfully demonstrated the world's first molecular transport system in molecular communication that uses molecules as an information medium. The envisioned applications of this molecular transport system are versatile in medicine and healthcare. This research was conducted jointly with the Sutoh Laboratory (Professor Kazuo Sutoh), Department of Life Sciences, The University of Tokyo and the Takeuchi Laboratory (Associate Professor Shoji Takeuchi), Institute of Industrial Science, The University of Tokyo.

Research Laboratories

Satoshi Hiyama
Yuki Moritani
Tatsuya Suda

1. Introduction

Molecular communication uses molecules (chemical substances such as proteins and deoxyribonucleic acids (DNAs)[*1]) as an information medium and is a new communication paradigm based on biochemical reactions caused by received molecules [1]-[4]. Since molecular communication was pioneered by NTT DOCOMO, it has received increasing attention as an interdisciplinary research area that spans the nanotechnology[*2], biotechnology[*3], and communication engineering. The National Science Foundation (NSF)[*4] has recognized the importance and impact of molecular communication research and has already started investigation into funding [5].

Molecular communication is inspired by the observation of biological molecular systems that living organisms have acquired through the evolutional process for more than billions of years. The communication in the biological systems is typically done through molecules (i.e., chemical signals). For instance, multicellular organisms including human beings perform maintenance of homeostasis, growth regulation, kinematic control, memory and learning through inter-cellular communication using signal-transducing molecules such as hormones [6]. Molecular communication aims to develop artificially designed and controllable systems that could transmit biochemical information such as phenomena and status of living organisms that is not feasible to transmit with the traditional communication that uses electromagnetic waves (i.e., electronic and optical signals) as an information medium.

Although molecular communication is slow speed, short ranged, and stochastic nature of communication, it has unique communication features including low energy-consumption, high compatibility with biological systems, and communicability in aqueous environment that are not seen in the traditional communication. Thus molecular communication is not competitive but complementary to the traditional communication.

General molecular communication

*1 **DNA**: A kind of nucleic acid that contains the genetic information of living organisms. This research uses artificially designed and synthesized DNAs that do not encode any genetic information for the purpose of constructing a molecular transport system and will not use biological genes in future years.

*2 **Nanotechnology**: A field of applied science whose theme is the control of matter on a nanometer scale. A nanometer equals to one millionth of a millimeter.

*3 **Biotechnology**: A field of biological engineering and technological application that uses living organisms, biological materials and information.

*4 **NSF**: A United States government agency that supports fundamental and innovative research in all the non-medical fields of science and engineering, resulting in producing over 100 Nobel Prize winners.

system includes a sender that emits information encoded molecules (called information molecules), a molecular communication interface that protects the emitted information molecules from denaturation (e.g., molecular deformation caused by changes in temperature or pH) in the propagation environment, a molecular transport system that transports the information molecules from a sender to a receiver, and a receiver that receives the transported information molecules and biochemically reacts to the received information molecules resulting in decoding of the information (**Figure 1**).

This article focuses on design of the molecular transport system that uses motor proteins[*5] driven by chemical energy[*6] and artificially synthesized DNAs, and explains experimental results that confirm the feasibility of the proposed system to transport specified molecules toward a designated site. In addition, this article describes that the molecular transport system would be applicable to a biochip[*7], or a biomolecule-analyzer, and presents an advanced health checkup service as an example medical/healthcare application using a mobile phone equipped with the biochip (called "biochip mobile phone"). This research has been conducted jointly with the Sutoh Laboratory and the Takeuchi Laboratory of The University of Tokyo, world leaders in the areas of motor proteins, DNAs, and microfabrication to construct the proposed system promptly.

2. Design of Molecular Transport System

The simplest and easiest approach to transport information molecules from a sender to a receiver is to use free diffusion. However, it is very difficult to realize a directional transport of information molecules due to the Brownian motion[*8] arising from frequent collisions of information molecules with disturbance molecules[*9] in the propagation environment.

In contrast, motor proteins such as kinesins[*10] directionally transport cargoes such as subcellular organelles and vesicles almost exactly to their destinations in spite of frequent collisions with disturbance molecules within a biological cell. This mechanism is known as an active transport and is realized by motor proteins' enzymatic actions that convert chemical energy called adenosine triphosphate (ATP) into mechanical work as directional walk along filamentous proteins such as microtubules (MTs)[*11] [6]. Motor proteins are nanometer-scaled actuators and have received increasing attention as engineering materials because they also work in the artificial environment outside of biological cells where aqueous conditions such as temperature and pH are reasonable [7]. For instance, directional gliding of MTs was demonstrated by immobilizing kinesins onto a glass substrate etched by lithography[*12] [8]. By applying this mechanism, we proposed a molecular transport system that uses gliding MTs as cargo transporters of information molecules. The idea is to load information molecules onto gliding MTs at a sender, to transport (glide) the information molecule-loaded MTs from a sender to a receiver, and to unload the transported information molecules from the gliding MTs at a receiv-

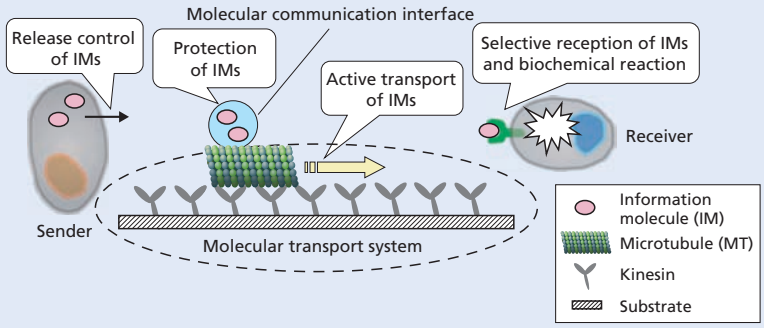

Figure 1 Molecular communication system

*5 **Motor protein**: A kind of protein that is capable of moving autonomously by converting chemical energy into mechanical work. For instance, it is responsible for producing the contractile force in muscle cells and is responsible for transporting cargoes in nerve cells.

*6 **Chemical energy**: A kind of energy that is produced by chemical reactions among molecules. In this article, it is used as fuel for motor proteins.

*7 **Biochip**: A fingertip-sized microchip packing traditionally bulky tools and reagents for biological and chemical experiments into smaller spaces. It is essentially miniaturized laboratories that can perform simultaneous chemical analysis and synthesis on the chip.

*8 **Brownian motion**: Random movement of particles such as molecules suspended in a liquid or gas due to the frequent collisions with medium molecules such as water molecules in water solution.

*9 **Disturbance molecule**: A group of molecules including water molecules, inorganic ions, organic small molecules, and organic macromolecules.

*10 **Kinesin**: A kind of motor protein whose size is several tens of nanometers. Typical kinesins are shaped like radish sprouts.

*11 **MT**: A cylindrical protein with approximately 25 nm-diameter and a few or tens of micrometers long.

er. Major issues to demonstrate this idea are how to load specified information molecules onto gliding MTs at a sender and how to unload the information molecules from the gliding MTs at a designated receiver in a reversible manner. To resolve these issues, we proposed a mechanism to load/unload information molecules using DNA hybridization[*13] between two single-stranded DNAs (ssDNAs)[*14] and strand exchange[*15] [9].

In order to use the DNA hybridization/strand exchange, each gliding MT, cargo (information molecule), and unloading site is labeled with different ssDNAs (**Figure 2** (a)). The ssDNA for the cargo is designed to be either complementary or non-complementary to that attached to the MT. When an MT labeled with an ssDNA passes through a given loading site (a given sender), a cargo labeled with an ssDNA complementary to that attached to the MT (cargo B) is selectively loaded onto the gliding MT through DNA hybridization (Fig. 2 (b)), while cargoes labeled with a non-complementary ssDNA (cargo A) remain at the loading site. Note that the length of an ssDNA attached to an MT is designed to be shorter than that attached to the cargo, and the long ssDNA attached to the cargo has single-stranded portion. This partial DNA hybridization is not as strong as the complete DNA hybridization and allows unloading of cargoes at an unloading site (a receiver).

The cargo loaded onto the MT (i.e., an MT-cargo complex) is transported by MT motility on kinesins toward given unloading sites (Fig. 2 (c)). Note that the length of an ssDNA attached to a cargo is designed to be as long as that attached to the unloading site, and the ssDNA attached to each unloading site is designed to be either complementary or non-complementary to that attached to the cargo. When the MT-cargo complex passes through an unloading site, the cargo labeled with an ssDNA complementary to that attached to the unloading site (unloading site B) is selectively unloaded from the gliding MT through strand exchange (Fig. 2 (d)). This unloading process through the strand exchange may be initiated by the energy state transition from an unstable and high-energy state (i.e., partial DNA hybridization between ssDNAs attached to the MT and to the cargo) to a stable and low-energy state (i.e., complete DNA hybridization between ssDNAs attached to the unloading site and to the cargo) that takes place naturally. Note that ssDNA-labeled MTs that unloaded cargoes continue to glide over immobilized kinesins and may load new cargoes.

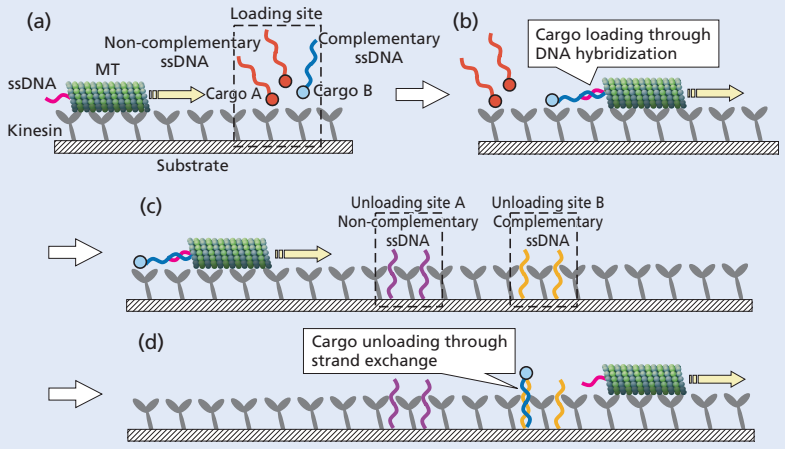

Figure 2 Mechanisms of a proposed molecular transport system

3. Experimental Results

In order to confirm the feasibility of the designed molecular transport system shown in Fig. 2, we firstly examined various chemical linkages to label MTs with ssDNAs and found that a chemical linkage that cross-links thiolated ssDNAs[*16] and amino groups of MTs was suitable for dense labeling of MTs with ssDNAs, while maintaining

*12 **Lithography**: A microfabrication technique used to make a micro-pattern through ultraviolet light or electron beam irradiation. Nowadays it is a core technology to make integrated circuits and microelectromechanical systems.

*13 **DNA hybridization**: A phenomenon that complementary base pairs (adenine and thymine, guanine and cytosine) of two single-stranded DNAs bind together to form a double-helix structure (double-stranded DNA) through hydrogen bonds.

*14 **ssDNA**: Strands whose all the base pairs in a double-stranded DNA melt and exist in solution as two entirely independent molecules. The corded, untwisted DNA strand is composed of combinations of the four bases (adenine, thymine, guanine and cytosine).

*15 **Strand exchange**: A phenomenon that upon melting a double-stranded DNA, one of the resulting single-stranded DNA and a third single-stranded DNA bind together to form a new double-stranded DNA.

*16 **Thiolated ssDNA**: A single-stranded DNA whose either or both of the asymmetric ends are modified with hydrogenated sulfur (thiol groups).

the smooth gliding of labeled MTs on kinesins [9].

Next we observed gliding MTs labeled with 10-base ssDNAs and cargoes (plastic microbeads with 1 μm-diameter) labeled with 23-base ssDNAs using microscopy. When an MT labeled with ssDNAs complementary to those attached to a cargo glided near a cargo (**Figures 3** (a), (b)), the cargo was occasionally loaded onto the gliding MT (Fig. 3 (c)). The resulting MT-cargo complex glided on the surface covered with kinesins to transport the loaded cargo (Figs. 3 (d), (e)) at an average speed of 0.34 μm/s [9]. Microscopic observation also confirmed that loaded cargoes were selectively unloaded from the gliding MTs at micro-arrayed unloading sites labeled with 23-base ssDNAs complementary to those attached to the cargo [10]. We confirmed that selective DNA hybridization/strand exchange was responsible for their cargo loading/unloading onto/from gliding MTs and the transport of the resulting MT-cargo complexes, respectively [9].

4. Example Medical/Healthcare Application

The successfully demonstrated molecular transport system delivers specified molecules to a designated site and it does not require external power supply or control as long as chemical energy exists in the aqueous propagation environment. Thus it is suitable for installation in small-sized devices, and could help lead to the realization of a biochip that operates in an autonomous manner.

The envisioned applications of the molecular transport system are versatile in medicine and healthcare. For instance, it may be possible to diagnose diseases or stress by directly analyzing biomolecules in a drop of sweat or blood using a "biochip mobile phone," a mobile phone equipped with a biochip (**Figure 4**). The molecular transport system would be packaged in the biochip that performs biochemical analysis, and the acquired results would be transmitted to a medical specialist via a mobile phone using traditional cellular networks. The system could be used, for example, for remote health checkups or preventive medicine. These advanced services are not science fiction but provide mobile daily health checkups that prevent some diseases before they occur or progress.

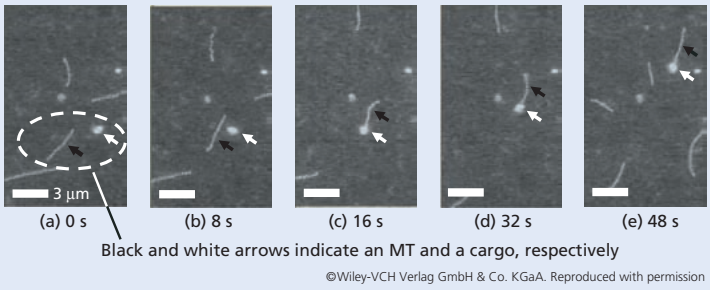

Figure 3 Microscopic images of cargo loading and transport

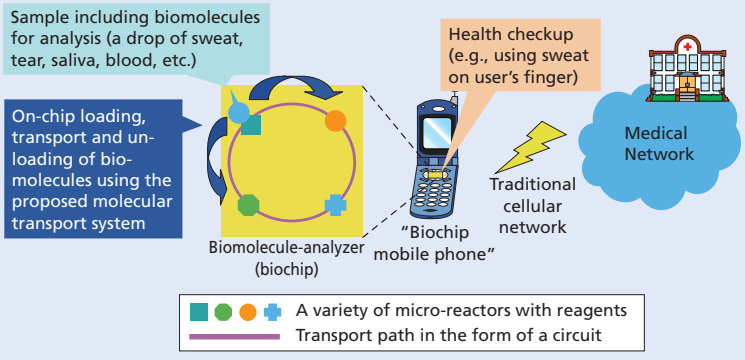

Figure 4 Envisioned mobile phone equipped with a biochip

5. Conclusion

This article focused on design and experimental results of a molecular transport system in molecular communication that uses motor proteins and DNAs to transport specified molecules toward a designated site. In addition, this article described a concept of a "biochip mobile phone" as an example medical/healthcare application of the proposed molecular transport system. NTT DOCOMO and The University of Tokyo are continuing their collaborative research into practical uses of molecular communication to identify applicable molecules and to develop an actual molecular transport system for installation in a biochip.

References

[1] S. Hiyama, Y. Moritani, T. Suda, R. Egashira, A. Enomoto, M. Moore and T. Nakano: "Molecular Communication," Proc. NSTI Nanotechnology Conference and Trade Show, Vol.3, pp.391-394, May 2005.

[2] S. Hiyama, Y. Moritani and T. Suda: "Molecular Communication," The Journal of the Institute of Electronics, Information and Communication Engineers (IEICE), Vol.89, No.2, pp.162-166, Feb. 2006 (In Japanese).

[3] Y. Moritani, S. Hiyama and T. Suda: "Molecular Communication — A Biochemically-Engineered Communication System — ," Proc. Frontiers in the Convergence of Bioscience and Information Technologies, pp.839-844, Oct. 2007.

[4] S. Hiyama, Y. Moritani and T. Suda: "An Overview of Molecular Communication and Its Medical Applications," Nanomedicine — Medical Applications of Nanotechnology — (Chapter 3-6), pp.248-258, Ohmsha Ltd., Feb. 2008 (In Japanese).

[5] NSF Workshop on Molecular Communication: Biological Communications Technology, Feb. 2008. http://netresearch.ics.uci.edu/mc/nsfws08/index.html

[6] B. Alberts, A. Johnson, M. Raff, P. Walter, D. Bray and K. Roberts: "Essential Cell Biology — An Introduction to the Molecular Biology of the Cell," Garland Publishing, Jul. 1997.

[7] M. G. L. van den Heuvel and C. Dekker: "Motor Proteins at Work for Nanotechnology," Science, Vol.317, pp.333-336, Jul. 2007.

[8] Y. Hiratsuka, T. Tada, K. Oiwa, T. Kanayama and T. Q. P. Uyeda: "Controlling the Direction of Kinesin-Driven Microtubule Movements along Micro-Lithographic Tracks," Biophysical Journal, Vol.81, No.3, pp.1555-1561, Sep. 2001.

[9] S. Hiyama, T. Inoue, T. Shima, Y. Moritani, T. Suda and K. Sutoh: "Autonomous Loading/Unloading and Transport of Specified Cargoes by Using DNA Hybridization and Biological Motor-Based Motility," Small, Vol.4, No.4, pp.410-415, Apr. 2008.

[10] S. Hiyama, S. Takeuchi, R. Gojo, T. Shima and K. Sutoh: "Biomolecular Motor-Based Cargo Transporters with Loading/Unloading Mechanisms on a Micro-Patterned DNA Array," Proc. IEEE International Conference on Micro Electro Mechanical Systems, pp.144-147, Jan. 2008.