



HOKKAIDO UNIVERSITY

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学位論文内容の要旨

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学位論文題名

Microtubule Swarm Programmed with *p-tert*-Butyl Substituted Azobenzene Tethered DNA
(*p-tert*-ブチル置換アゾベンゼン修飾 DNA によってプログラムされた微小管の集団運動)

Swarming is a collective behaviour displayed by moving or self-propelled objects purely by local interactions among them. In nature, a wide range of self-propelled objects such as animals, birds, cells, and bacteria exhibit swarming or other collective behaviours to construct fascinating large scale patterns with emergent functions. Inspired by these attractive features, various approaches to understand and demonstrate swarming based on engineered systems such as synthetic, chemical, and robotic systems have been reported until now. Alongside with these approaches, natural biomolecular motor systems, microtubule (MT)-kinesin have emerged as ideal candidates for experimentally demonstrating collective behaviours, due to the small size and self-propelled ability. However, it was difficult to control the local interactions for the swarming among the self-propelled biomolecular motors. To overcome this problem, photoresponsive DNA (*p*DNA) conjugated MT was developed. Photo-regulated swarming of MTs were achieved previously by employing DNA which contains azobenzene as the photoswitch where the formation and dissociation of the swarm in response to Vis and UV light, respectively (Vis-ON swarm system). However, having only one type of photoswitch to control the swarming could potentially become a bottleneck, when trying to construct a further complicated system. In this dissertation, the main objective was to establish a new photo-regulated biomolecular motor-based swarm system for the expanded application in the field of nanotechnology.

In **chapter 1**, the purpose of this dissertation and the background of this study are described.

In **chapter 2**, swarming of MTs after UV light irradiation and the dissociation into individual MTs upon visible light irradiation is demonstrated. This control of swarming is entirely reversed from the previously reported system where swarming was initiated with visible light irradiation and the dissociation upon UV light irradiation. In addition, the effect of UV light irradiation

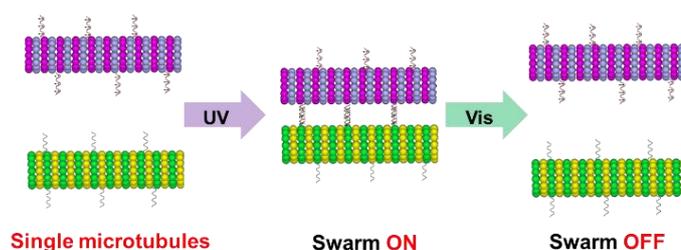


Figure 1. Scheme of swarm formation and dissociation

towards the swarm formation was systematically investigated as well as the control of reversible swarming of MTs in a repeated manner by irradiating the MTs with alternate visible and UV light irradiation.

In **chapter 3**, the novel behaviours of photoresponsive DNA carrying *para tert*-butyl azobenzene that was used for the photo-regulation of swarm under UV light irradiation is demonstrated by carrying out standard fluorescent quenching experiment and UV-Vis absorption measurement to evaluate a ratio of duplex formation. At the photo-stationary state (PSS) of UV light, the DNA formed the duplex with the complimentary DNA strand due to the *trans*-to-*cis* isomerisation of the azobenzene. However, unexpected discovery of the partial dissociation of the duplex into the single strands by the prolonged UV light irradiation after PSS was observed. In addition, when the sample was incubated in dark after the UV light irradiation, it induced the duplex formation.

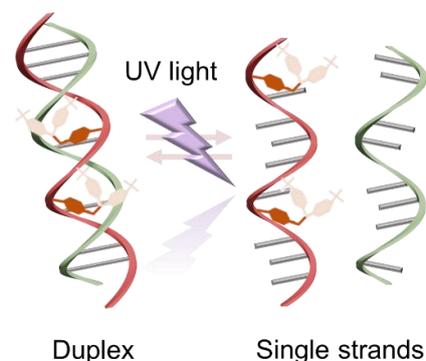


Figure 2. Scheme of possible behaviour of DNA duplex under the UV light irradiation.

In **chapter 4**, an orthogonal control of swarm system is achieved by integrating UV-ON and Vis-ON swarm systems in parallel to the same experimental system. The swarms were actively responded by associating and dissociating into individual MTs in response to the appropriate light signal. Until now, orthogonal, and reversible control was only demonstrated in static systems, hence this is the first report on the orthogonal control of the self-propelled biomolecular motor system where MTs can actively respond to the different light signals by emerging swarms or dissociating into individuals.

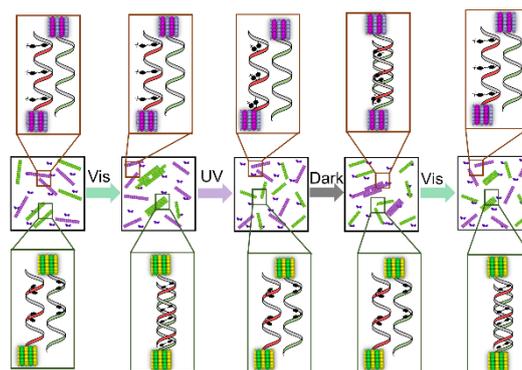


Figure 3. Scheme of orthogonal control of swarming in response to a light signal.

In **chapter 5**, all the important results and future aspects of this research are summarised as the concluding remarks.

This dissertation describes the establish a new photo-regulated biomolecular motor-based swarm system, where *para tert*-butyl azobenzene was incorporated into the DNA backbone as a photoswitch to regulate ON/OFF switching of the swarming. Introducing a new photoswitch incorporated DNA extend the programmability of the biomolecular motor system to expand the potential applications of biomolecular motors in many fields ranging from biotechnology to developing photo-regulated molecular machines. Furthermore, the approach of introducing different photoswitches in the same system may contribute to the construction of further complex swarm systems for extended applications in molecular robotics.