A novel single-microparticle total analysis system driven by optical tweezers

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Microparticles in the environment play key roles in transportation of chemically or biologically toxic substances because of their high adsorption ability and high mobility. For instance, microplastics (1-5000 μm plastic particles) can adsorb toxic chemicals such as persistent organic pollutants (POPs) and heavy metals and convey them to wild creatures. While methods to analyze surface chemicals of such microparticles are needed for detailed estimation of their risks, current known ones are very limited.

In this research, we propose a novel system of analyzing individual microparticles with active manipulation by optical tweezers. In this system, sample microparticles are adaptively manipulated by optical tweezers, then analyzed in detail by touching their multiple surface points sequentially to the objects called "analysis ports," which provide various functions such as Raman spectroscopy, fluorescence spectroscopy, and biosensing. The advantage of the proposed system is that it enables multiple chemical analyses of the bulk and the surface of individual particles. In this paper, we demonstrated the feasibility of the proposed system by realizing manipulation of non-spherical polystyrene microparticles (about 50 μm) with optical tweezers and detection of fluorescent paint attached to the surface of the particles using an analysis port of localized light illumination.

1. Introduction

Microparticles in the environment are thought to have a risk of acting as transporters of pollutants because they adsorb chemically or biologically toxic substances and they are light enough to be scattered in the environment (Fig. 1). Especially, pollution caused by microplastics (particles of $1-5000\ \mu m$ made from plastics[1]) are currently regarded as one of the main environmental problems. They can do harm to creatures which ingest them since microplastics can sorb (adsorb or absorb) persistent organic pollutants (POPs), inorganic pollutants (e.g., heavy metals), and pathogens[1]. A study showed that organic pollutants adsorbed by plastic pellets in seawater were 10^5-10^6 times as much as the ones in bulk seawater[2]. Besides microplastics, atmospheric aerosol particles such as mineral dust can also adsorb POPs and transport them over long distances[3]. Despite the presence of potential risks, detailed behavior of the adsorption on environmental microparticles are still not known well.

Therefore, in order to estimate their risks, analysis methods for adsorbed pollutants on microparticles are highly needed. Currently, they are usually analyzed by extraction from many particles and gas chromatography[4]. However, this method is not adequate for the estimation since it lacks the ability to treat each particle individually to investigate pollutants adsorption on the whole surface. Such individual analyses are essential because microparticles in the environment vary in the sense of many parameters such as size, shape, colors, and material, and relations of these parameters are still not unclear. A study using TOF-SIMS partially realized this individual analysis[5], but it was limited to the detection of elements, which is not suitable for organic pollutants.

To solve this problem, we propose a novel concept of a system which analyzes microparticles (Fig. 2). We call this system the single-microparticle total analysis system driven by optical tweezers. This system handles each microparticle individually using optical tweezers

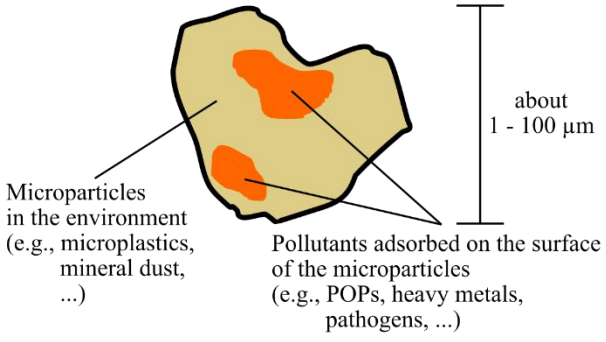


Fig. 1 Environmental microparticles focused on in this research.

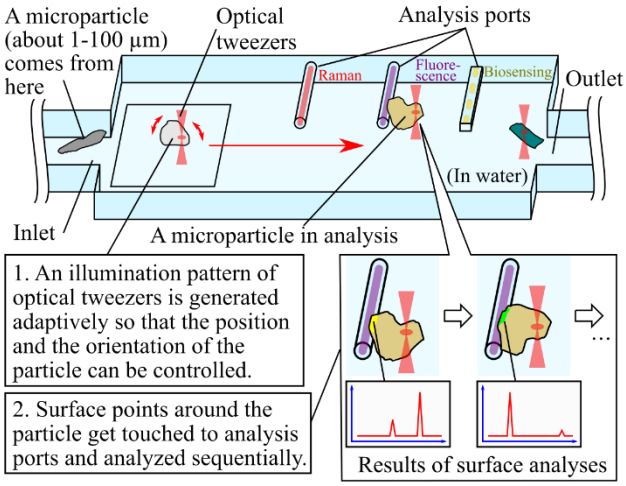


Fig. 2 The concept of the single-microparticle total analysis system driven by optical tweezers.

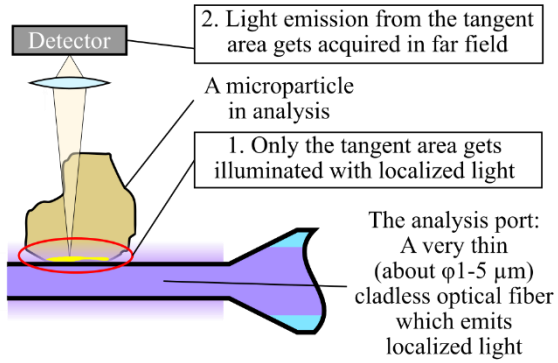


Fig. 3 The analysis port of localized light illumination.

technique[6], a tool to trap microparticles (usually dielectric ones) with optical radiation force, and produces a result of a total analysis (i.e., a set of integrated results about its materials and adsorbed pollutants on it). The targets of this system are mainly microparticles of about 1 – 100 μm, for which further analyses are especially needed in the case of microplastics[1]. The advantage of the proposed system is that it enables multiple chemical analyses of the bulk and the surface of individual particles.

In this paper, we report on the realization of the single-microparticle total analysis system driven by optical tweezers with fundamental settings. We also report on a verification experiment of the selective surface analysis conducted with the system. The selective surface analysis means to analyze only a part of the surface of microparticle which is touched to the analysis port. It is one of the most important functions of the analysis system since it is required for analyses of the whole surface of microparticles. In the experiment, we conducted detection of fluorescent paint on a non-spherical polystyrene microparticle (about 50 μm), with the detection area confined to the tangent area between the microparticle and the analysis port, using manipulation with optical tweezers and analyses with an analysis port of localized light illumination. We conducted this experiment in order to show the feasibility of the concept, realizing the selective surface analysis, which is one of the most important functions of analyses in the system.

2. Proposal of the single-microparticle total analysis system driven by optical tweezers

2.1 The main concept of the system

Fig. 2 shows the basic idea of the system. The steps of the analysis with this system are as follows. When a microparticle comes in from the inlet, the camera captures its image, and the system starts tracking its motion. Next, the particle is optically manipulated by the trapping laser (it acts as optical tweezers), whose illumination pattern is adaptively generated using the captured image. Then, it gets transported to an area with what are called “analysis ports.” Each of the analysis ports conducts some analysis method such as Raman spectroscopy, fluorescence spectroscopy, or biosensing. When a microparticle is touched to the analysis ports, they can selectively analyze the area of its surface which is tangent to the ports. With these ports, by controlling the position and orientation of the particle using optical tweezers to touch the points around the particle surface to the ports sequentially, data of the whole surface get collected. Finally, the collected data get integrated into a detailed result of a total analysis of the whole surface of the particle.

Among many tasks required for realization of the concept, the following two are the main ones:

Task 1.

To establish a way to manipulate microparticles of arbitrary shape using optical tweezers.

Task 2.

To realize analysis ports that can selectively analyze the surface area of microparticles which is tangent to the ports.

In the experiment of this paper, we tackled these two tasks with basic settings. For Task 1, we demonstrated trapping of a non-spherical polystyrene microparticle (about 50 μm) to the analysis port using optical tweezers. This case, for simplicity, the illumination pattern of optical tweezers was just one point. For Task 2, we demonstrated detection of fluorescent paint attached to the particle using an analysis port of localized light illumination (details are described in Section 2.2).

2.2 Implementation of an analysis port of localized light illumination

In the proposed system, as an example of the selective surface analysis, we implemented an analysis port of localized light illumination (Fig. 3). This port is made of a very thin (about 1 – 5 μm in diameter) cladless optical fiber that can emit localized light (evanescent light) when light is introduced. It acts the same as tapered optical fibers[7], which are widely used for sensing. This port has a function of illuminating the tangent area of the particle with localized light.

In this paper, we conducted an experiment of detecting fluorescent paint attached to the surface of non-spherical polystyrene microparticles (about 50 μm) using this analysis port. In this experiment, the function of the port is the same as total internal reflection fluorescence (TIRF) microscopy[8]. When a particle touches the port, only the tangent area of the particle gets illuminated by the localized light. Then, fluorescent substances in that area get excited to emit fluorescent light. Finally, by observing the particle with

microscope, fluorescent substances only on the tangent area of the particle can be detected selectively.

3. Experimental system

In this paper, we developed an experimental system, which is a simple realization of the single-microparticle total analysis system driven by optical tweezers. The schematic diagram of the experiment system is shown in Fig. 4(a). The device was composed of an optical tweezers unit, a microscopy unit, an excitation laser unit, a controller unit, and a sample cell. The trapping laser used in the optical tweezers unit was an infrared one (976 nm, 0 − 350 mW) whose output power could be controlled manually. The optical tweezers unit was equipped with 2D Galvano mirrors, which could control the focus (i.e., the point of illumination) of the trapping laser horizontally. The objective lens of the optical tweezers unit was shared with the microscopy unit so that the trapped particle can be observed through the cameras. The microscopy unit had a color camera, a monochromatic camera, and an LED lighting system. Between the camera and the objective lens, a long-pass filter which cut out the light from excitation laser could be inserted. The excitation laser unit could introduce 405 nm laser into the analysis port. We call this laser the excitation laser because fluorescence of paint was excited using light from this laser in the experiment. The control unit included a PC, which can control the 3D stepper motor stage, 2D Galvano mirrors, and cameras. In the PC, a graphical application developed for this experiment system was used to capture the color camera images and to order the points of illumination manually in accordance with the pixels of the camera images. The sample cell was equipped with an analysis port of localized light illumination, which is fabricated by etching one end of an optical fiber made from glass (Fig. 4(b)). The suspension of sample microparticles was injected into the sample cell.

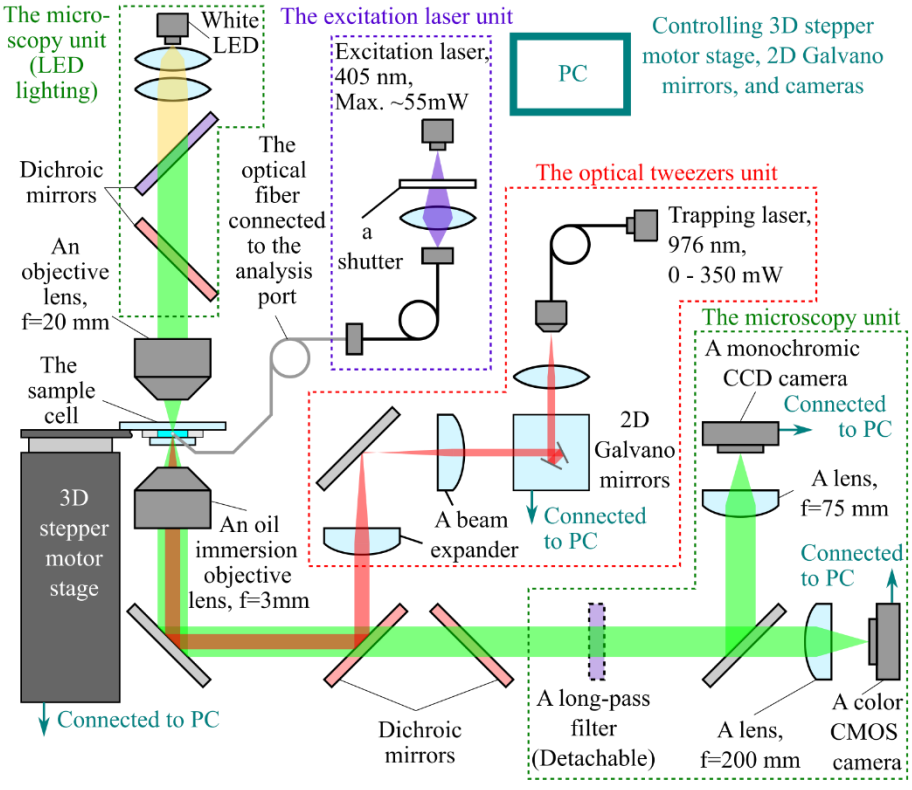
4. The verification experiment of the selective surface analysis

4.1 Preparation of sample microparticles

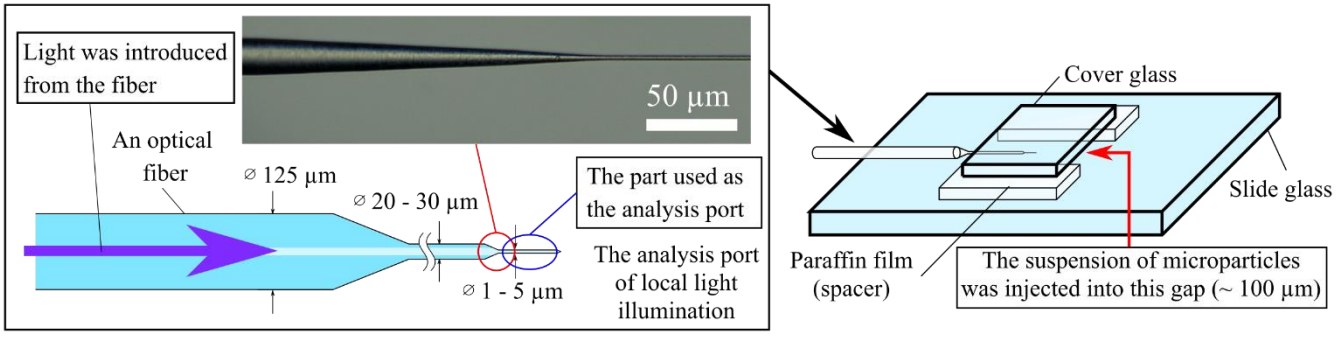
In this experiment, fluorescent paint was employed as an example of pollutants adsorbed on the surface of microparticles. Non-spherical polystyrene microparticles partially coated with fluorescent paint were collected as dust made in the process of polishing a polystyrene spoon coated with fluorescent paint with a file. The particles were collected in water solution of a surfactant (Approx. 0.65 % solution of sodium dodecyl sulfate). The surfactant was used in order to improve dispersion stability. Typical sample particles are shown in Fig. 5(a).

4.2 Experiment schemes

Fig. 5(b) shows the scheme of the experiments. First, the particle with fluorescent paint was trapped using optical tweezers and transported to touch the analysis port by moving the cell relatively to the focal point using the 3D stepper motor stage. Then, light of the excitation laser was introduced into the analysis port, and the particle was illuminated with the induced localized light. Finally, the presence of

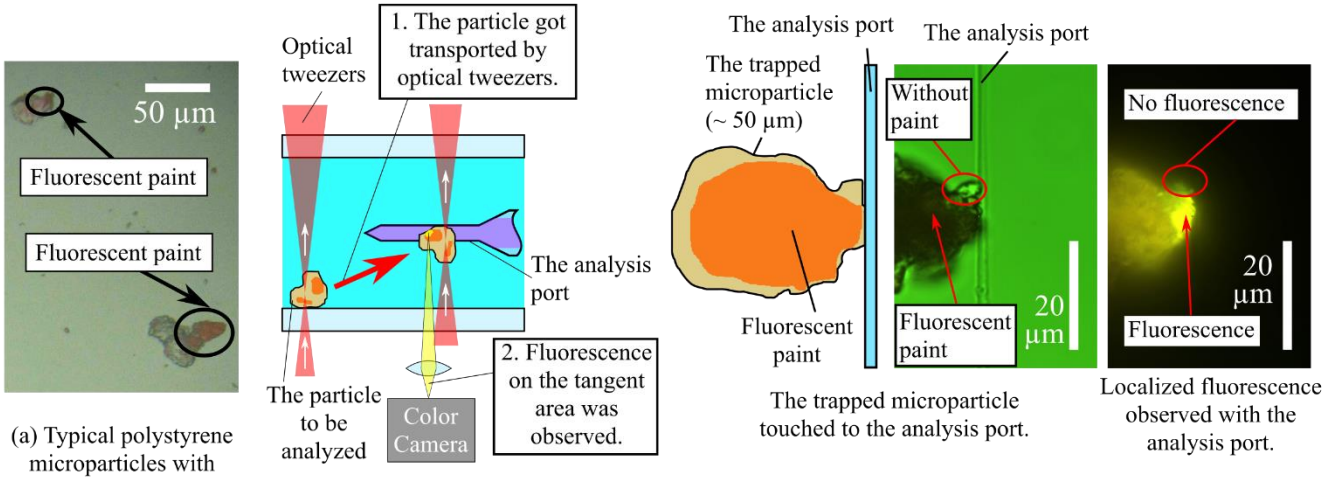


(a) The schematic diagram of the developed system.



(b) The structure of the sample cell.

Fig. 4 The system developed for the verification experiment of the selective surface analysis.



(a) Typical polystyrene microparticles with fluorescent paint. (b) The experiment schemes. (c) The results of the experiment.

Fig. 5 The verification experiment of the selective surface analysis.

fluorescence from the tangent area was observed with the color camera.

4.3 Results and Discussion

Fig. 5(c) shows the results of the experiment. In the experiment, a non-spherical polystyrene microparticle (about 50 μm) with fluorescent paint on its surface was trapped to touch to the analysis port using optical tweezers (The output was about 20mW), and when illuminated with the excitation laser, yellow light from a part of the tangent area was observed. This part was in accordance with the area covered with fluorescent paint. Moreover, when the LED light for transparent illumination was turned off and the long-pass filter was inserted to cut off the excitation light for the observed microscopic image, this yellow light was still observed in the camera. This result shows that this yellow light was fluorescent light from the paint. In addition, the emission of this yellow light was confined to the tangent area between the particle and the analysis port. This indicates that fluorescence was excited selectivelys in the tangent area.

Therefore, we can conclude that the selective surface analysis was successfully demonstrated with the detection of substances attached to the surface of non-spherical microparticles (about 50 μm), with the detection area confined to the tangent area between the microparticle and the analysis port, using an analysis port of localized light illumination.

5. Conclusion

In this paper, we propose a concept of the single-microparticle total analysis system driven by optical tweezers, a novel system of analyzing individual microparticles to investigate their materials and adsorbed pollutants on them with active manipulation by optical tweezers. In addition, we developed an experimental system, which is a basic realization of the single-microparticle total analysis system driven by optical tweezers. Moreover, we conducted a verification experiment of the selective surface analysis with this system. In the experiment, we conducted detection of fluorescent paint on a non-spherical polystyrene microparticle (about 50 μm), with the detection area confined to the tangent area between the microparticle and the analysis port, using manipulation with optical tweezers and analyses with an analysis port of localized light illumination. The results show the feasibility of the concept of the single-microparticle total analysis system driven by optical tweezers, realizing the selective surface analysis, which is one of the most important functions of analyses in the system. The single-microparticle total analysis system driven by optical tweezers is a tool that can realize far more detailed investigation about microparticles and their adsorbed pollutants than current methods, which contributes to further research on transportation phenomena of pollutants caused by microparticles in the environment.

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