

# Dynamical Micro-Bead Pattern Forming Using Laser Manipulation Techniques

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**Abstract** — Micro-bead arrays/patterns have potential for sensing tools in various scientific fields such as medicine, biochemistry, and so on. Laser trapping is an eminently suitable tool for dynamical handling of micro-beads. Here we demonstrate the fully automated assembly of micro-bead patterns by the control of an isolated-point-type laser trapping field in time domain. Several tens of micro-beads automatically form the specified patterns from random initial positions, and subsequent metamorphoses succeed.

## 1. Introduction

DNA chips using micro-spots of bio-molecules on a glass slide are valuable tools in genomics. Dynamic micro-arrays/patterns using mobile substrates, usually micro-beads coated with bio-molecules/chemicals, offer greater flexibility and have the potential to be used as sensing tools in not only scientific fields such as genomics and molecular biology but also engineering and medical fields such as lab-on-a-chips, drug discovery, diagnostics, and so on [1]. In the several demonstrated methods, laser manipulation techniques, namely optical tweezers, are eminently suitable for dynamic handling of micro-beads, since they have several advantages [2].

In this paper, we demonstrate the fully automated assembly of micro-bead patterns by the control of an isolated-point-type laser trapping field in time domain, that is, optical tweezers based on intelligent control techniques. First, we mention the principle of optical tweezers, and outline the experimental setup for the demonstrations. Secondly, we describe the algorithm for dynamical micro-bead patterning. Finally, the experimental results using the three-beam laser manipulation system demonstrate the performance of our approach.

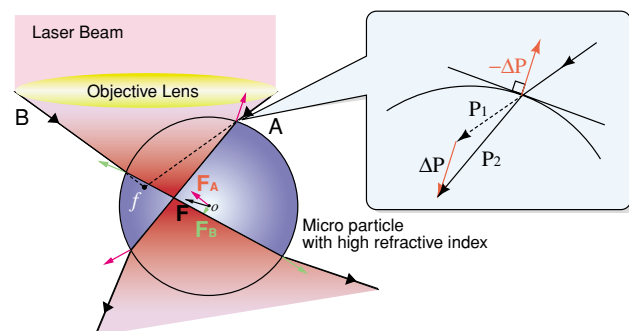
## 2. Laser Manipulation

### 2.1. Principle

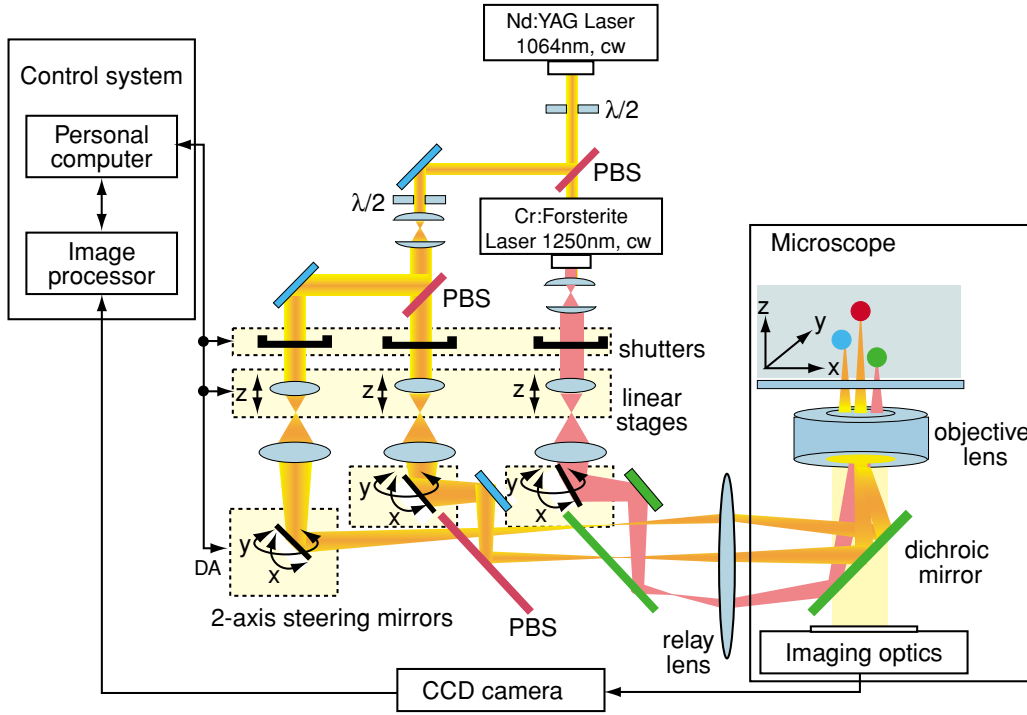
Laser trapping, first demonstrated in 1970 by Ashkin [3] and well known as optical tweezers [4], has been further extended to line-scanning [5], holographic [6], time-sharing [7], and others. Many of optical tweezers techniques except for generalized phase contrast (GPC) [8] are derived from strongly focused laser beams. Conventional optical tweezers, which have an isolated-point-type trapping field, use forces exerted by a strongly focused laser beam to trap small objects ranging in size from sub-micro-meters to several tens micro-meters. The basic principles are straightforward for objects much smaller than the wavelength of light, or much larger [4]. Larger object acts as lens, refracting the rays of light and redirecting their photons' momentum,  $\Delta P$ , shown schematically in Fig. 1. The resulting recoil draws them toward the higher flux of photons near the focus. For example, in Fig. 1, the refraction of a pair of rays A and B of the trapping beam gives forces  $F_A$  and  $F_B$  whose vector sum  $F$  is always restoring for axial and transverse displacements of the particle from the trap focus  $f$  [9].

### 2.2. Experimental System

Time-sharing scanning methods of laser trap are useful for testing new control algorithms of the dynamic micro-



**Figure 1:** Principle of the laser trapping of a micro particle.  $F$ : trapping force,  $f$ : focal point of a laser beam,  $\Delta P$ : change of photons' momentum.



**Figure 2:** Three-beam time-sharing synchronized scanning optical tweezers system.  $\lambda/2$ : half-wave retardation plate, PBS: polarizing beam splitter.

bead patterns, since they are simple to rapidly change multiple trapping positions. The laser scanning methods with high-reflective mirrors also enable the use of powerful trapping beams. Hence, we have developed a three-beam Time-Sharing Synchronized Scanning (3B-T3S) optical tweezers system, which can translate the patterns to the XY-plane at the desired Z-coordinate. Figure 2 shows the optical configuration and control system of the 3B-T3S optical tweezers system. Our 3B-T3S system is configured around an inverted microscope with high-NA oil-immersion objectives. The laser trapping sources are a continuous wave (CW) Nd:YAG laser (1064nm, 16W(max)) and a CW Cr:Forsterite laser (1250 nm, 800mW(max)), and the former laser beam is split into s- and p-polarized beams by polarizers. The powers of these beams are adjusted by half-wave retardation plates. The three trapping beams are introduced coaxially into the microscope via shutters, lenses, 2-axis steering mirrors, a relay lens and the fluorescence port, and they are reflected upward by a dichroic mirror to the objective. The time-shared focal positions on the XY-plane are controlled by the 2-axis steering mirrors. The Z-coordinate of each time-shared beam is also controlled by the lens mounted on each PC-controlled linear stage which can move parallel to the optical axis. An image processor (Hitachi IP5005) digitizes the images (512×512 pixels) from a color CCD camera in real time for feature recognition. Software developed for the image processing and device control is executed by a personal computer (PC).

### 3. Dynamic Micro-bead Pattern

#### 3.1. Control Algorithm

Dynamical micro-bead patternings and their transformation require the complex coordinated movements of a large number of beads. The movements are controlled with multi-beam optical tweezers under the bead detection and path generation algorithm. The Hough technique, which is the most common algorithm known to be robust under noisy conditions such as microscope images, is applied to the bead detection. The trajectories for parallel transportation without collisions are gradually generated by the following algorithm. First, the algorithm assigns pre-designed destinations,  ${}^d\mathbf{p}_k$ , to initial positions,  ${}^i\mathbf{p}_k$ , which are detected by the circular Hough transform. Second, new positions in the next time-step are gradually updated by

$$\mathbf{p}_k(t_{s+1}) = \mathbf{p}_k(t_s + \tau) = \mathbf{p}_k(t_s) + \delta\mathbf{p}_k, \quad s = 0, \dots, n-1, \quad (1)$$

where  $\delta\mathbf{p}_k$  is the step size vector of  $k$ -th bead,  $\mathbf{p}_k(t_0) = {}^i\mathbf{p}_k$ , and  $\mathbf{p}_k(t_n) = {}^d\mathbf{p}_k$ . Since we have found that an optimal step size for smooth transportation using optical tweezers techniques is less than the radius of the bead, each  $\delta\mathbf{p}_k$  is determined by the following equations:

$$\delta\mathbf{p}_k = [\delta x_k, \delta y_k]^T = ({}^d\mathbf{p}_k - {}^i\mathbf{p}_k) / n, \quad (2)$$

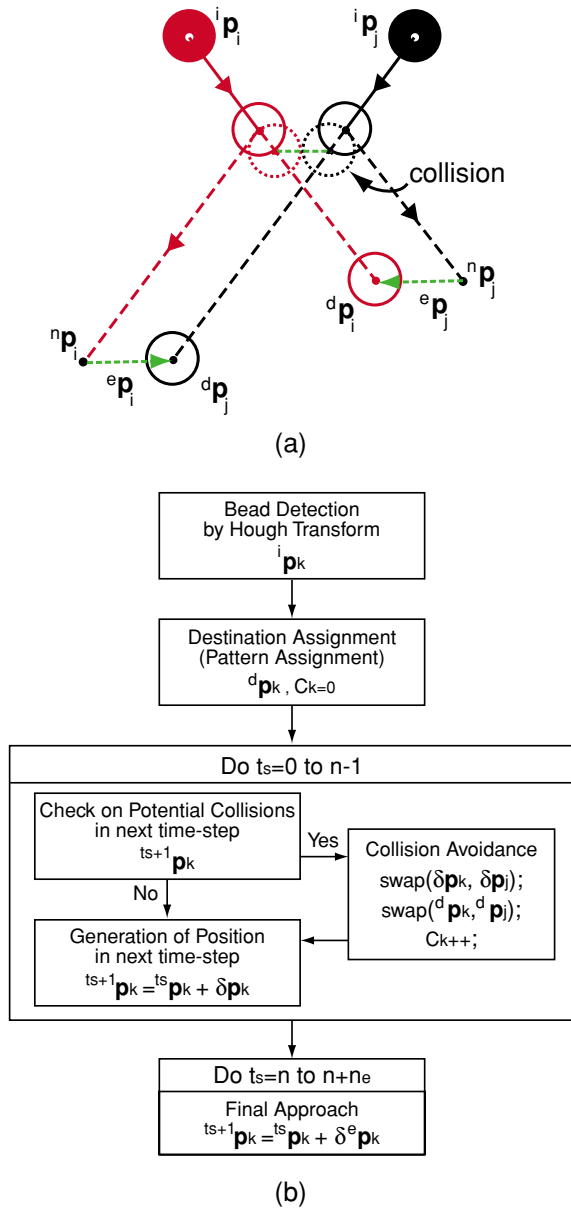
$$n \geq \max \left| \frac{{}^d\mathbf{p}_k - {}^i\mathbf{p}_k}{r} \right|, \quad k = 1, \dots, M, \quad (3)$$

where  $r$  is the radius of the bead,  $M$  is the number of destinations, and  $n$  is the smallest integer satisfying the Eq. (3). Under Eqs. (1)-(3), particle collisions cannot be avoided,

since each bead is transported directory toward its destination along the linear trajectory. Therefore, we have adopted a rule for avoiding collisions; if a collision occurs in the next time-step, we simply exchange both the step size vectors and the destinations as shown in Fig. 3(a), and add one to the  $k$ -th collision counter  $c_k$ . If once collision occurs, the beads corresponding to this collision cannot reach their destinations at  $t_s = n$ , that is,  ${}^n\mathbf{p}_k \neq {}^d\mathbf{p}_k$ . Finally, therefore, the procedure for final approach to their destinations is necessary if the collision occurs. The positions for this final approach are updated by

$$\mathbf{p}_k(t_{s+1}) = \mathbf{p}_k(t_s + \tau) = \mathbf{p}_k(t_s) + \delta^e \mathbf{p}_k, \quad s = n, \dots, n + n_e, \quad (4)$$

$$\delta^e \mathbf{p}_k = ({}^d\mathbf{p}_k - {}^n\mathbf{p}_k) / n_e, \quad (5)$$



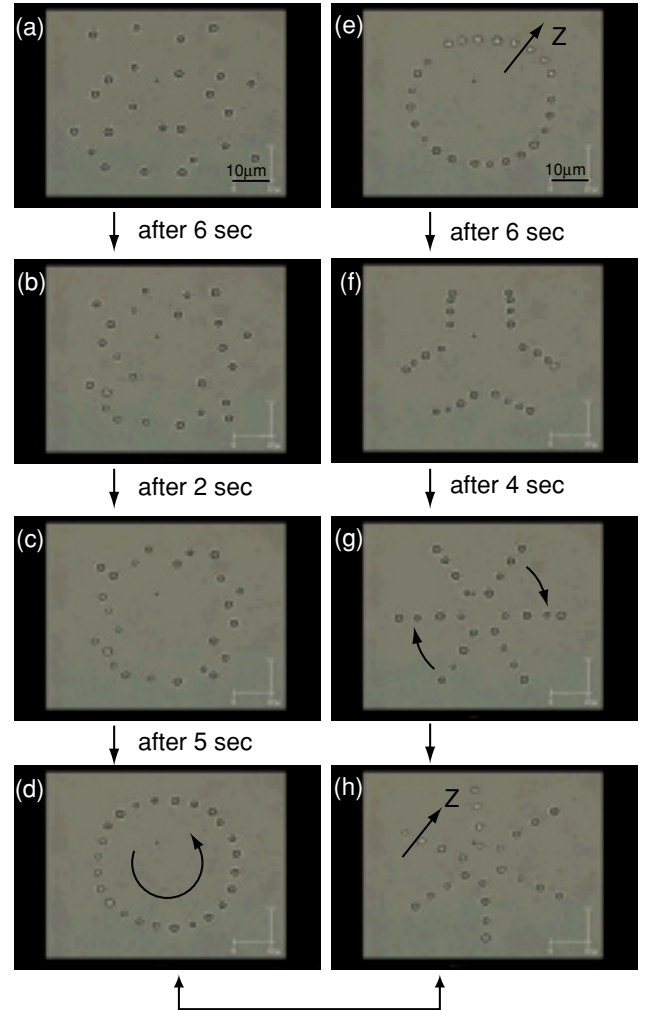
**Figure 3:** Algorithm for dynamical micro-bead patterning without collisions. (a): Schematic of collision avoiding, (b) diagram of updating bead positions.

$$n_e = \max \left| \frac{{}^d\mathbf{p}_k - {}^n\mathbf{p}_k}{r} \right| < 4 \max(c_k), \quad k = 1, \dots, M, \quad (6)$$

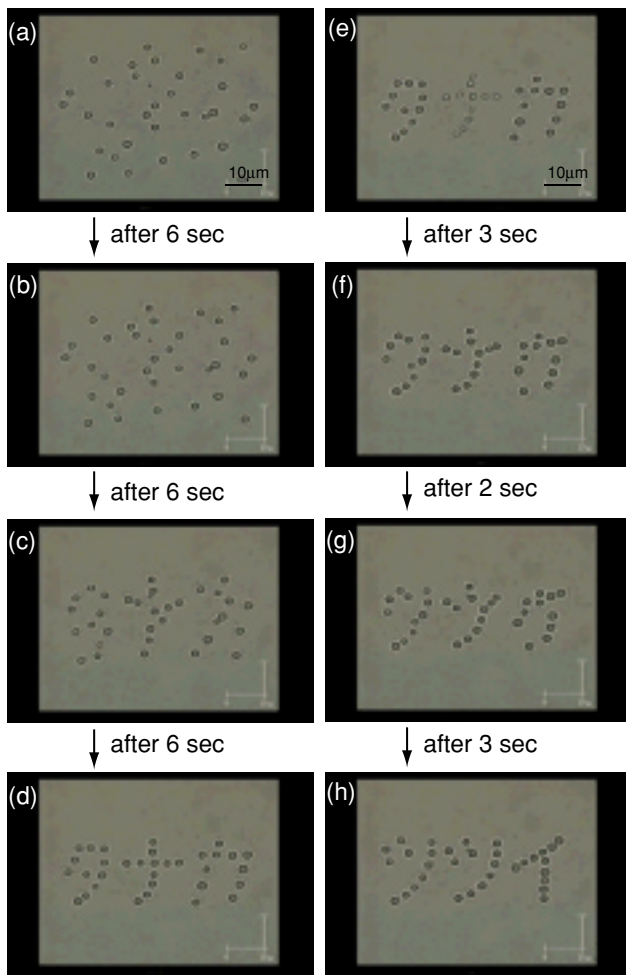
where  $\delta^e \mathbf{p}_k$  is the step size vector for the final approach, and  $n_e$  is the smallest integer satisfying the Eq. (6). The diagram of the algorithm is shown in Fig. 3(b). Note that the adequate assignment of destinations is important for less potential collisions. The assignment method for lattice pattern is proposed in our previous paper [2].

### 3.2. Demonstrations

On the basis of the control algorithm mentioned in the above section, we demonstrated the fully automated dynamical patterning of micro-beads. The sample was glass spheres (borosilicate, 2.5 $\mu\text{m}$ ), and the beam powers were adjusted to roughly 200mW (beam1, beam2, and beam3 were 205mW, 212mW, and 156mW, respectively). The scanning dwell time for stable trapping was also adjusted to 0.01 seconds for each bead. Figure 4 shows a sequence of images recorded with the CCD camera showing the result of automated assembly of a dynamic bead array with specified pattern and its metamorphoses. First, the center positions of micro-beads dispersed in water droplets on the



**Figure 4:** Video frame sequence of dynamical forming of a pattern and its metamorphoses.



**Figure 5:** Video frame sequence of dynamical forming of Japanese characters and their metamorphoses.

cover glass were detected by the circular Hough transform, then all twenty-four beads in the field of view were, automatically and simultaneously, trapped at the initially detected positions using 3B-T3S optical tweezers (Fig. 4(a)). Second, the twenty-four trapped beads were simultaneously transported to pre-designed destinations where three sets of eight beads formed a circle, while avoiding collisions each other (Fig. 4(b)-4(c)). Third, after the circle pattern was rotated in a counterclockwise direction (Fig. 4(d)), each set of eight beads was translated to Z-coordinate direction (Fig. 4(e)). Fourth, the circle was transformed to a stellar pattern (Fig. 4(e)-4(g)). Finally, the stellar pattern was transformed again to a circle after the rotation and translation (Fig. 4(h)).

In another demonstration shown in Fig. 5, thirty-three beads in the pipetted droplets were automatically patterned to form the Japanese character 'katakana'. First, each laser beam which configured one T3S optical tweezers automatically trapped the set of eleven beads, and each set of eleven beads formed one character in the katakana syllabary, where three syllabics meant first author's name 'tanaka' (Fig. 5(a)-5(d)). Second, syllabics were sequentially translated to Z-coordinate direction in order of 'ta'- 'na'- 'ka' (Fig. 5(e)). Finally, the pattern was transformed to the

different syllabics which meant second author's name 'tsutsui' (Fig. 5(h)), while avoiding collisions between each bead. Figures 5(f) and 5(g) show the intermediate status of beads through the transforming.

#### 4. Conclusion

We have demonstrated the fully automated assembly of micro-bead patterns by the control of an isolated-point-type laser trapping field in time domain. Our approach can be applied to not only micro-beads but also biological materials and colloidal structures. Furthermore, the dynamical forming of micro-bead arrays with not only lattice [2] but also specified, arbitrary patterns may be valuable sensing tools for lab-on-a-chips and be actuators for micro-fluidic devices.

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