

DVD Pick-up Based Optical Detection for Diffusive Mixing in Microchannels

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Abstract

Microfluidics deals with behavior, precise control, and manipulation of fluids at a micro scale. It has become increasingly prevalent in various applications, such as biomedical applications, inkjet heads, fuel cells, etc. The issue of mixing in microfluidic channels is often crucial in design, fabrication, and detection of microfluidic devices. In this paper, we propose a new optical method to characterize diffusive mixing in two types of microchannels (Y-type microchannel and premixer gradient generator). In order to measure the mixing of fluidic streams, we use an optical method using a DVD pick-up module, which is widely employed in optical storage systems installed within PCs or laptops. By injecting fluorescent dye into a solution, we measure the fluorescent intensities across the microchannels in order to visualize and quantify diffusive mixing. Experimental micrographs are compared with computational fluid dynamics (CFD) simulations. We show that our proposed optical scanner can be used as an alternative measurement system with high performance and cost-effectiveness in comparison with conventional optical tools such as epifluorescent microscopes and confocal microscopes.

Keywords: Microfluidics, Optical pick-up, Optical scanner, Microchannel, Diffusive mixing

Introduction

Microfluidics has become increasingly prevalent and has found widespread use in various applications, such as biomedical applications (diagnostics, therapeutics, and cell/tissue engineering)¹⁻³, inkjet heads⁴,

and fuel cells⁵. Because of their scaled-down size, microfluidic devices have significant advantages, including lower consumption of reagents, fast response, increased automation, and reduced manufacturing costs⁶. However, reduced analytes in microfluidics and the corresponding reduction in detection volumes make it difficult to detect measurable properties of the analytes. For example, electrochemical detection, based on conductivity and potential difference, may not fulfill its own function in microfluidic devices because of such downscaling⁷. On the other hand, fluorescence detection has been used successfully with microfluidic devices⁸ because of its superior selectivity and sensitivity on a small scale³. Nowadays, epifluorescent microscopes with high-resolution CCD cameras and confocal microscopes with photomultiplier tubes (PMT) are generally used to detect and observe microfluidic devices. However, because these devices are costly and lack portability, we need cost-effective and easily accessible platforms for use with microfluidic devices.

Some researchers have used optical storage technology as the detection mechanism of choice for the fluorescence-based DNA chip. CD technology has been proposed to build a detection system with equivalent or even better performance than conventional scanners⁹. Furthermore, a lab-on-a-chip (LOC) system has been developed for application in DNA analysis using a CD disk^{10,11}. Recently, we developed an optical scanner adopting a DVD pick-up module used in commercial DVD drives and showed its feasibility for fluorescence measurement in DNA chips, thereby achieving low cost, high sensitivity, and compact size¹². In this paper, we extend our previous work on the DVD pick-up based optical measurement to characterize microfluidic mixing in microchannels. Using fluorescent dye, we measure the fluorescent intensity quantitatively that represents fluidic patterns and the performance of diffusive mixing in the Y-microchannel and gradient-generating microchannel and compare these experimental results with simulated computational fluid dynamics (CFD) results.

Results and Discussion

Fluorescent Detection in Microchannels

We first measured diffusive mixing for the fabricat-

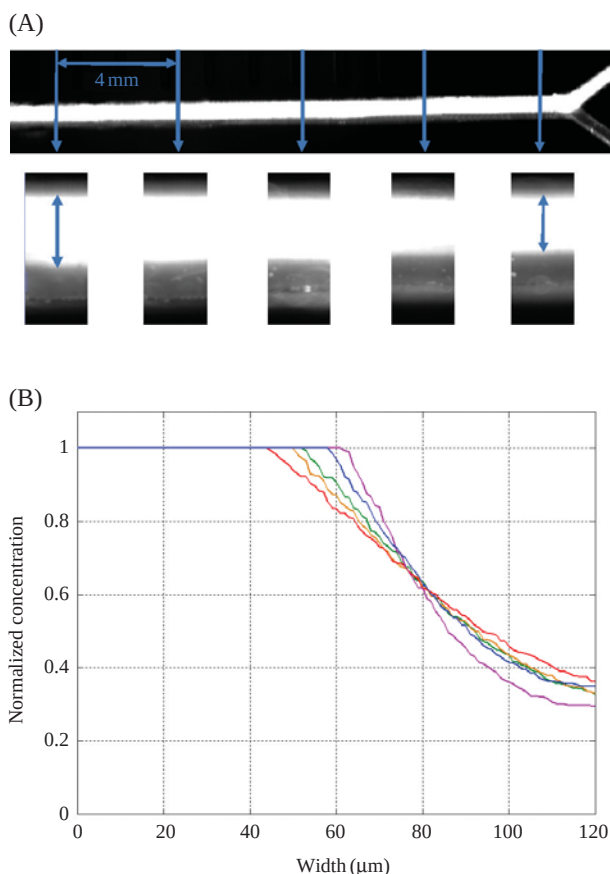


Figure 1. Experimental results for diffusive mixing in Y-microchannel. (A) Overview scanned fluorescent image in 10 μm mode and detail image in 1 μm mode. (B) Concentration profiles using intensities acquired from fluorescent images.

ed Y-microchannel using our proposed optical system. Deionized (DI) water was introduced to one input channel, and diluted Dylight 680 was introduced to the other input channel with the same flow rate in the microfluidic device; then, the mixture of the two flows was observed using our proposed scanner. The overview image of the Y-channel was taken in the 10 μm resolution mode, and detailed images were captured in the 1 μm resolution mode with 4 mm intervals, as shown in Figure 1(A). Linear gradient diffusions of fluorescent dye were successfully displayed, and fluorescent intensities were evaluated from captured images using image J. Gradients from the maximum normalized intensity value ($C_0=1$) to the minimum value ($C_0=0$) became smooth as the position of observation moved to the output of the microchannel, as shown in Figure 1(B). Theoretically, the fluorescent intensity of pure DI water can be assumed to be zero, but DI water actually has some fluorescent intensity, and the experimental results also showed that mini-

mum intensity in the Y-channel converged on a non-zero value.

We also carried out similar experiments for a fabricated premixer gradient generator with Christmas tree structures. The gradient-generating microchannel utilizes laminar flow and diffusive mixing to generate a continuous combinatorial mixture of two different solutions^{13,14}. Figure 2 shows scanned fluorescent images and micrographs in the fabricated gradient-generating microchannel. We measured continuous profiles of fluorescence intensity by diffusive mixing at four different points across the width (2,500 μm) of the microchannel. As with the case of the Y-microchannel, the DVD pick-up based optical device can successfully probe gradient generation by diffusive mixing in the Christmas tree microchannel. Typical stair-like fluorescent patterns in the premixer gradient generator could be detected in main channel, and these stair patterns proceed smoothly as output to the main channel.

Comparison of Experimental Results with Simulation

The Y-channel is a T-type mixer and simply combines two or more fluid streams that flow parallel to each other in the microchannel, and mixing relies purely on molecular diffusion. Many good and comprehensive experimental and numerical studies have been conducted for the Y-channel¹⁵⁻¹⁷; therefore, this microchannel was selected as an ideal model to evaluate and compare the performance of our proposed system.

We have used the commercial numerical tool FEM Lab (Comsol Inc., USA) for simulation of the Y-channel. The inlet flow is laminar and fully developed, with an average velocity of 350 $\mu\text{m}/\text{sec}$. A constant reference pressure of 0 Pa is set at the outlet. The equations for the momentum balance are the stationary Navier-Stokes equations with no-slip wall conditions:

$$-\nabla \cdot \eta(\nabla \mathbf{u} + (\nabla \mathbf{u})^T) + \rho(\mathbf{u} \cdot \nabla)\mathbf{u} + \nabla p = 0 \quad (1)$$

$$\nabla \cdot \mathbf{u} = 0$$

where η denotes the dynamic viscosity ($\text{kg}/(\text{m} \cdot \text{s})$), $\mathbf{u}$ is the velocity vector (m/s), ρ represents the fluid's density (kg/m^3), and p denotes the pressure (Pa). The fluid's properties are not affected by the change in concentration of the dissolved species.

The mass flux is given by diffusion and convection, and the resulting mass balance is as follows:

$$\nabla \cdot (-D\nabla c + c\mathbf{u}) = 0 \quad (2)$$

where D denotes the diffusion coefficient (m^2/s) and c is the concentration (mol/m^3). In this simulation, we used $D=0.84 \times 10^{-9} \text{ m}^2/\text{s}$ for ethanol¹⁸, because fluorescent dyes are very small molecules and the diffusion

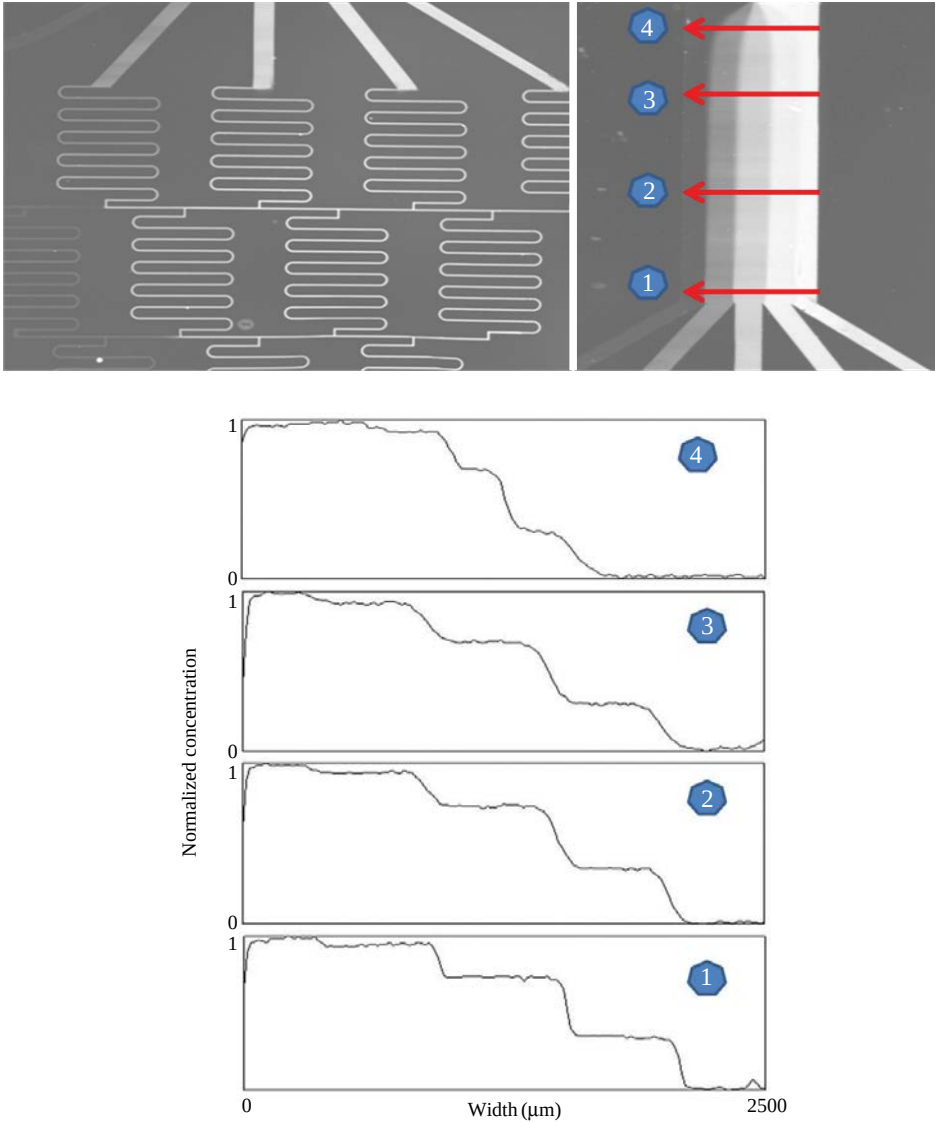


Figure 2. Scanned fluorescent images and intensity profiles for premixer gradient generator using proposed scanning system.

coefficients for typical small molecules are around $1 \times 10^{-9} \text{ m}^2/\text{s}$. At the outlet of the microchannel, the mass transport is mainly driven by convection; that is, the transport by diffusion is neglected in the normal direction of the cross section of the microchannel.

Figure 3 describes the multiphysics simulation results for this microchannel, taking into account the diffusion effect. We assumed that high concentration medium ($c=1$) and pure DI water ($c=0$) flowed in the Y-channel, and the distributions of concentration were profiled according to the length of the microchannel in the diffusion region, with each 4-mm interval as in the case of our experiments. The mixing occurred on only the interfaces between two flows because of the characteristics of laminar flow, and more molecules were diffused as the measurement

point moved to the end of the microchannel. When comparing the experimental results in Figure 1(B), the simulation results showed good agreement except that there are non-zero intensities at DI water regions in the experiments. From these comparisons, the proposed system showed its potential as a measurement system to analyze diffusive mixing quantitatively.

Discussion

From our experimental and simulation results, we showed the feasibility of using the proposed system to analyze diffusive mixing patterns in the microchannel quantitatively with the intensity of the fluorescent signal. However, it is very difficult to compare the analytical performance of the proposed system for microfluidics with the analytical performance of the com-

mercial confocal microscope directly, because there are many different parameters, such as optical path, properties of the objective lens, existence of a pinhole, etc. Therefore, in this study, we intended to compare the performance of the two systems in an indirect and simple manner. We defined a quality factor (Q.F.) as the numerical aperture of the objective lens times the sensitivity of the PMT detector

$$Q.F. = NA \times \text{Sensitivity.} \quad (3)$$

We chose a commercial confocal microscope from

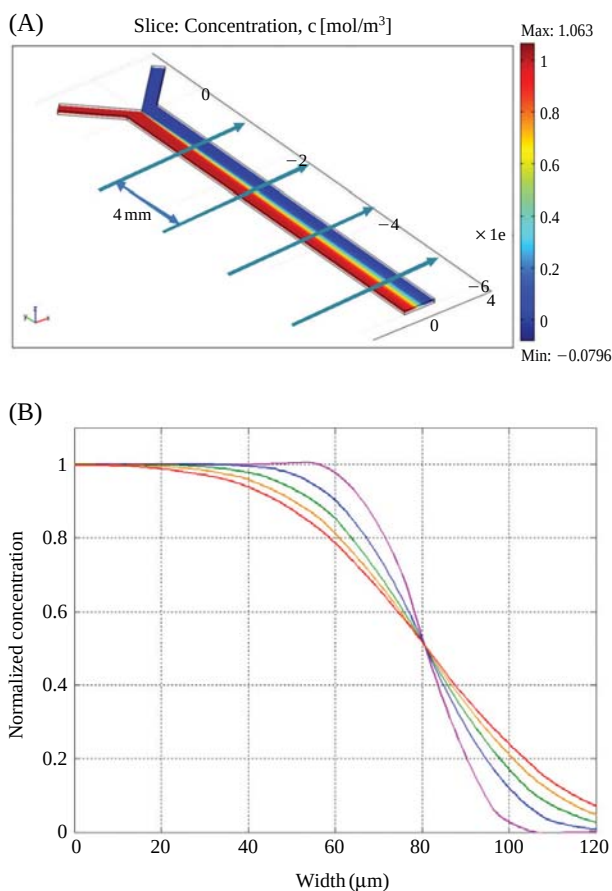


Figure 3. Simulation results for diffusive mixing in Y-microchannel. (A) Distributions of concentration using multiphysics simulation. (B) Concentration profiles from simulation results.

Carl Zeiss, Inc. (LSM 510, Germany), employed a PMT detector, and constructed a table (see Table 1) to compare the quality factors. Upon comparing these parameters, we determined that the Q.F. of our proposed system is between the minimum and maximum values obtained from a confocal microscope, as shown in Table 1. Therefore, we have demonstrated that our proposed system is as effective as the commercial confocal microscope.

Conclusions

We proposed an optical detection method to characterize diffusive mixing in a microchannel, based on DVD optical pick-up. We demonstrated that a low-cost optical scanner can be used effectively to examine microfluidic characteristics, and that it may be a cost-effective alternative to expensive epifluorescent microscopes. By comparing the concentration profiles of the FEM Lab simulation with experimental results using the standard diffusion microchannels, the optical system showed acceptable performance for detection of diffusive mixing, in line with the performance of the confocal microscope. Recently, Blu-ray disk (BD) and HD-DVD drives have been marketed by major optical disk drives companies as next-generation high-density optical data storage devices that can store several hours of HDTV movies. Therefore, we can easily extend current fluorescent detection and measurement techniques for characterizing microfluidic devices to those employing Blu-ray optical pick-up.

Materials and Methods

Optical Scanner Using a DVD Pick-up

Figure 4(A) shows a schematic diagram of the optical paths of a proposed optical scanner based on a DVD optical pick-up. It uses two laser diodes of 650 nm (red) and 532 nm (green) wavelengths for the detection of Cy3 and Cy5 fluorescent dyes, respectively. Because the DVD pick-up module includes only a 650 nm laser diode, we added a commercial green la-

Table 1. Comparison between quality factors (Q.F.) of optical scanner and confocal microscope.

	Laser sources/ Objective lens	NA	Sensitivity (A/W)	Q.F.
Optical scanner based on DVD pickup	Red laser (650 nm)	0.64	18,000	11,520
	Green laser (532 nm)	0.66	18,000	11,880
Confocal microscope	1.5 × O.L	0.35	24,000	8,400
	10 × O.L	0.55	24,000	13,200
	20 × O.L	0.8	24,000	19,200

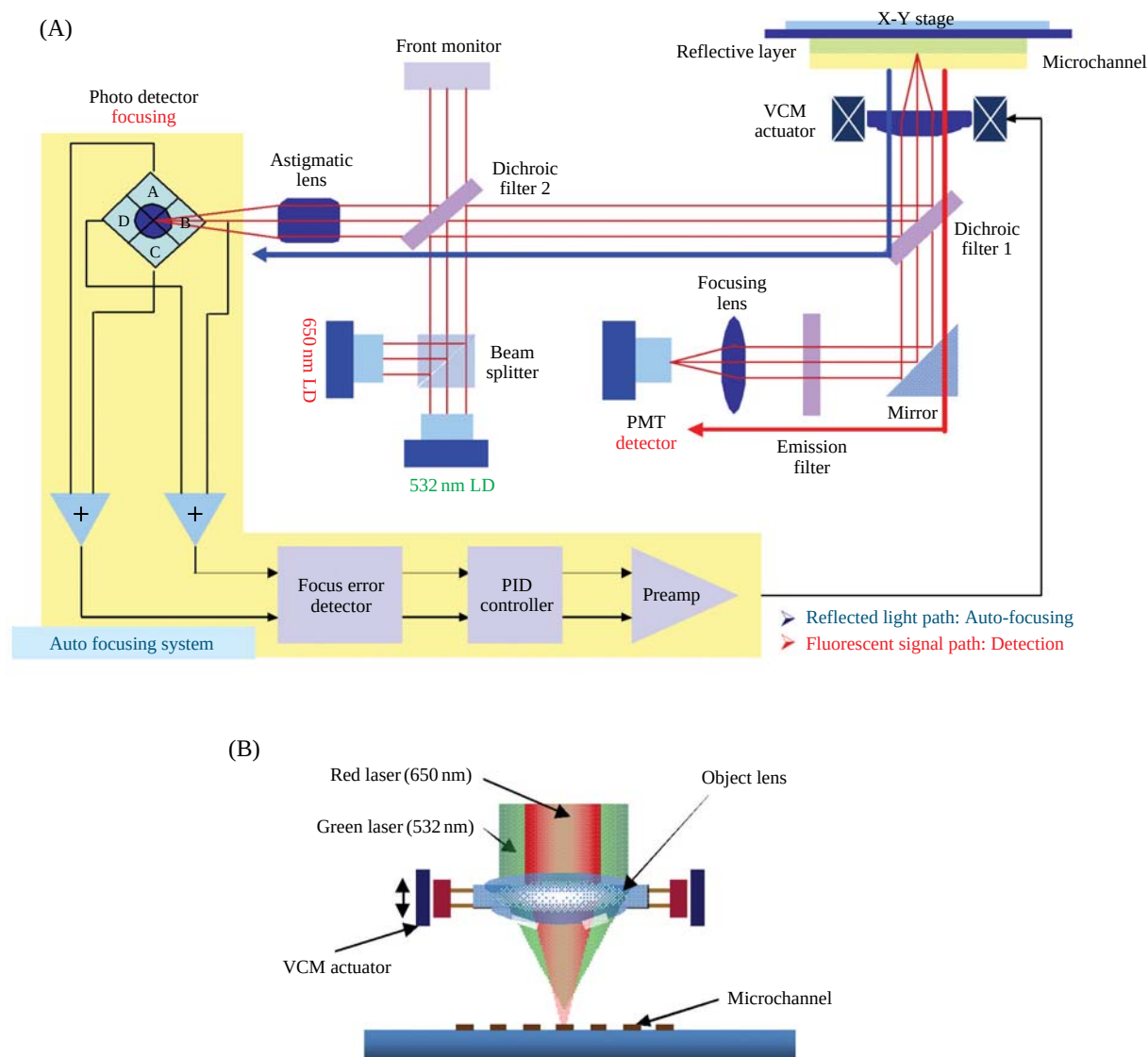


Figure 4. (A) Schematic diagram of optical path. (B) Two laser beams focused on microchannel surface through DVD objective lens.

ser diode into the system. There are two different optical paths; one is for auto-focusing and the other is for fluorescent detection. One of the laser diodes generates a laser beam of wavelength 650 nm or 532 nm. Most of the radiation from the laser beam is reflected by a dichroic filter designed for transmission of the two laser wavelengths; the part of the laser beam radiation passing through the dichroic filter is used to regulate the laser power by a front monitor sensor. The laser beam radiation transmitted through the dichroic filter is focused on the reflective layer, and the reflected beam is finally focused onto a four-quadrant photodiode (PD) after passing through two dichroic filters. The focus

error signal is calculated by using the intensities of the four divided regions of the PD. The PD generates a focus error signal whose magnitude is dependent upon the distribution of the beam spot across its four divided regions. The resulting servo signal is used to drive the VCM actuator in such a way that the objective lens is shifted to a point where its focal point falls upon the spot center. The laser beam radiation that is focused on the microchannel surface excites Cy5 dye (or Cy 3) in solution, thereby causing the dye to emit a fluorescent signal of 670 nm (or 570 nm). The collimated emission beam is then refocused by a detection lens and passes through a pinhole into the photomultiplier tube

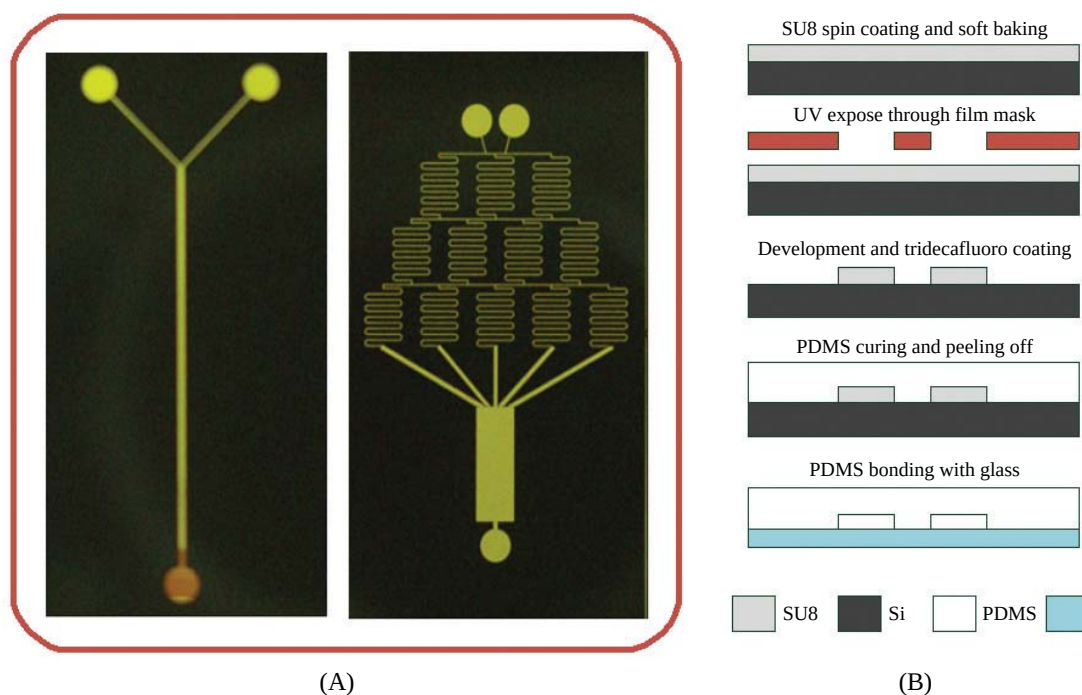


Figure 5. (A) Schematics of Y-type microchannel and premixer gradient generator (Christmas tree microchannel). (B) Fabrication process for microfluidic devices.

(PMT). The analog electric signal from the PMT represents the intensity of the fluorescent signal.

Figure 4(B) shows the DVD objective lens with a numerical aperture (NA) of 0.64 and the lens actuator. Both red and green laser beams can be focused on the microchannel surface through the objective lens. The emitted fluorescence and the reflective light from microfluidic devices are detected by the photomultiplier tube to obtain high detection sensitivity, and a dynamic autofocus mechanism compensates for any aberrations.

Fabrication Procedures for Microfluidic Devices

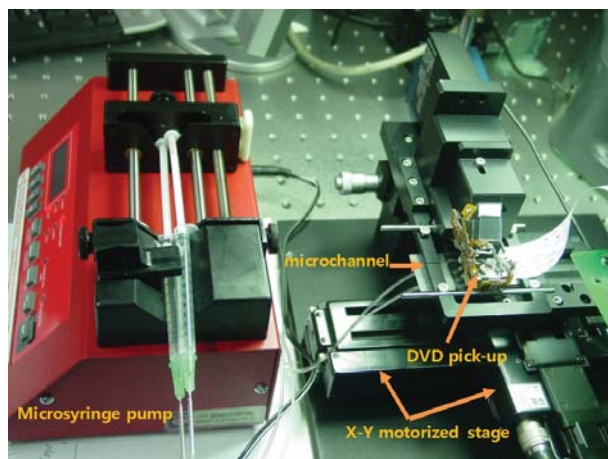
Figure 5 shows schematic diagrams that describe procedures for fabricating microfluidic devices. We fabricated a Y-shaped microchannel with 500 μm width, 50 μm height, and 30 mm length and a premixer gradient generator with Christmas tree structures using soft lithography to investigate the performance of our fluorescent scanner. A new silicon wafer was spin-coated with negative photoresist (SU-8, MicroChem Inc.), and the coated silicon wafer was soft-baked for several minutes. The wafer was then exposed under the mask using the aligner and was placed on a hot plate for several minutes of post-exposure baking, followed by a short relaxation time. Post-exposure baking

was followed by development at room temperature, and the whole wafer was rinsed with isopropyl alcohol (IPA) to clean residues from the wafer. A PDMS precursor (Sylgard 184 Silicone Elastomer, Dow Corning) and a curing agent were mixed at the ratio of 10 to 1, based on weight. Before the PDMS mixture was poured onto the fabricated master, the master was silanized with (tridecafluoro-1,1,2,2-tetrahydrooctyl)-1-trichlorosilane (Sigma Chemical Co., St. Louis, MO, USA) to allow easier removal of the PDMS after curing. The PDMS mixture was poured onto the master and cured at 80°C for 2 hrs. Then, the cured PDMS channel was peeled off from the master, cut and punched to connect microtubes, and bonded with a slide glass after O_2 plasma treatments.

Image Analysis and Other Experimental Setup

In this study, we used DyLight 680 dye (Thermo Scientific, USA), which corresponds to Cy 5.5 dye, in order to observe fluorescent intensity in the microchannel. Fluorescent intensities can be obtained directly from the electrical signal in the PMT detector or can be assessed from the captured images quantitatively using Image J (NIH, USA) software. A precise micro-syringe pump (NE-1000, New Era Pump Systems, USA) was employed to control the flow rate in the

(A)



(B)

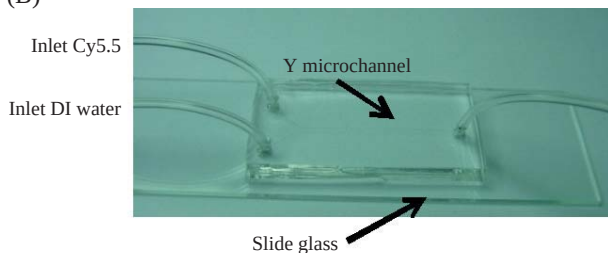


Figure 6. (A) Experimental setup for optical scanner using a DVD pick-up. (B) Fabricated microchannel located on the slide glass.

microchannel through the microtubes. Figure 6(A) shows the experimental system based on the DVD pick-up module with two laser sources. Figure 6(B) shows the fabricated Y-type microchannel attached to the glass slide. The fabricated microfluidic devices are installed on precise X-Y linear stages in a 1 μm or 10 μm resolution mode.

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References

1. Brivio, M. *et al.* Miniaturized continuous flow reaction

- vessels: Influence on chemical reactions. *Lab on a Chip* **6**, 329-344 (2006).
2. Dittrich, P.S. & Manz, A. Lab-on-a-chip: Microfluidics in drug discovery. *Nature Reviews Drug Discovery* **5**, 210-218 (2006).
3. Reyes, D.R. *et al.* Micro total analysis systems. 1. Introduction, Theory, and Technology. *Analytical Chemistry* **74**, 2623-2636 (2002).
4. Petersen, K.E. Fabrication of an integrated, planar silicon ink-jet structure. *IEEE Transactions on Electron Devices* **ED-26**, 1918-1920 (1979).
5. Yamazaki, Y. Application of MEMS technology to micro fuel cells. *Electrochimica Acta* **50**, 663-666 (2004).
6. Becker, H. & Locascio, L.E. Polymer microfluidic devices. *Talanta* **56**, 267-287 (2002).
7. Kuswandi, B. *et al.* Optical sensing systems for microfluidic devices: A review. *Analytica Chimica Acta* **601**, 141-155 (2007).
8. Whitesides, G.M. The origins and the future of microfluidics. *Nature* **442**, 368-373 (2006).
9. Perraut, F. *et al.* A new generation of scanners for DNA chips. *Biosensors & Bioelectronics* **17**, 803-813 (2002).
10. Kido, H. *et al.* Disc-based immunoassay microarrays. *Analytica Chimica Acta* **411**, 1-11 (2000).
11. Matsui, T. & Shimonura, T. DVD optical-head with fluorescent detection. Optical data storage topical meeting (2006).
12. Kim, K. *et al.* A new DNA chip detection mechanism using optical pick-up actuator. *Microsystem Technologies* **13**, 1359-1369 (2007).
13. Dertinger, S.K.W. *et al.* Generation of gradients having complex shapes using microfluidic networks. *Analytical Chemistry* **73**, 1240-1246 (2001).
14. Lin, F. *et al.* Generation of dynamic temporal and spatial concentration gradients using microfluidic devices. *Lab on a Chip* **4**, 164-167 (2004).
15. Kamholz, A.E. *et al.* Quantitative analysis of molecular interaction in a microfluidic channel: The T-sensor. *Analytical Chemistry* **71**, 5340-5347 (1999).
16. Kamholz, A.E. & Yager, P. Molecular diffusive scaling laws in pressure-driven microfluidic channels: deviation from one-dimensional Einstein approximations. *Sensors and Actuators B-Chemical* **82**, 117-121 (2002).
17. Ismagilov, R.F. *et al.* Experimental and theoretical scaling laws for transverse diffusive broadening in two-phase laminar flows in microchannels. *Applied Physics Letters* **76**, 2376-2378 (2000).
18. Crussler, E.L. Diffusion Mass Transfer in Fluid Systems. New York, Cambridge University Press (1984).