Flow cell detection with an optical fiber

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ABSTRACT

In this experiment, we tested various optical fibers to select a suitable fiber for a effective flow cell detection. In order to align between micronozzle and optical fibers, a guide channel was fabricated by Si wafer etching with MEMS (Micro Electro-Mechanical System) technology. This fabricated system is advantageous due to its low cost and simple structure. This is possible because it replaces many optical lenses and expensive equipment with optical fibers. In result of cells detection by using various fiber modes in this system, for multi mode both input and output part, it was easy to align. The sensitivity of cell detection was not bad as other cases. The output voltage was as high as about 300_, so we are going to use a light source which has relatively small output power. Therefore, it was suitable for minimization. By injecting various cells, we were able to detect cells. The reason for the various voltage characteristics is the difference of light permeability for each cell which is made up of specific cellulose and cell walls. In addition, we were able to compare the relative size of the injected cells.

Keywords: Cytometric system, Optical fiber, Flow cell detection

1 INTRODUCTION

It has made an effort of applying optical fibers to the commucication. The light for a communication can send more information than an electric wave. The reason is the light has higher frequncy than an electric wave. An optical fiber is used to the endoscope, and all sorts of detector for pressure and temperature and so on. The optical fiber which is made of synthetic resins, is used to the bright decoration of flower shaped. Among various applications, it is used to the detection of flow cell for this thesis.

Flow cytometry is the system measuring the optical signal obtained from flowing cells uniformly after it applies a laser light to cells. That is, a lot of biological phenomena in cells are measured by real time accessing. And it is classified the cells by the biological ingredients and structure. So it is certainly the essential equipment studying on life science such as medical science, biology, genetics, etc. But it is difficult to apply in these studies due to high cost, big size and complex mechanism.

Therefore we miniaturized the established flow cytometry with the optical method and MEMS technology. We have fabricated the micro flow cytometric system with advantage of low cost for fabrication and maintenance. And it is a simple structure and use because it replaces optical fibers by many optical lenses and expensive equipments.

2 FABRICATION

The principle of proposed system is that transfers also the optical signal, output signal of micro nozzle to electical circuit by the optical fiber after focusing the laser light on micro nozzle by optical fiber. Then AD conversing, it measures and analysis the signal by real time data acquisition system, the designed and fabricated circuits.

2.1 Optical fiber

The optical fiber is composed of core, cladding and coating. The core is in the center of the fiber and is the medium of propagation for the optical signal. The core is made of silica glass or plastic with a high refractive index. Typical core sizes range from 8 microns for single mode silica glass cores up to 1000 microns for multi mode plastic optical fiber. And the cladding is a material of lower index of refraction which surrounds the core. This difference in index forms a mirror at the boundary of the core and cladding. Because of the lower index, it reflects the light back into the center of the core, forming an optical wave guide. That is. the refractive index of a core is higher than the refractive index of a cladding. A light is focused to a core and progressed without a leak. The optical fiber which core size is several microns is called by single mode optical fiber and tens of microns is called by multi mode optical fiber. For single mode, the loss is small but it is difficult to align among the optical fibers. Then it is used to a long-distance trasmission and a big-capacity communication. Multi mode which is used to short-distance transmission is suitable for detection of flow cells. Because a core size is big at tens of microns, the output signal is big also. It is able to apply a light source with smaller output power relatively. It is going to fit at a miniature.

2.2 Fabrication

In order to align between optical fibers and micro nozzle, the guide channel is fabricated of (100) silicon wafer with MEMS technology. Figure 1 represents the fabricated micro nozzle. The Si wafer was etched by TMAH(20.0 wt%, 90_, etch rate: 0.97 _/min). To

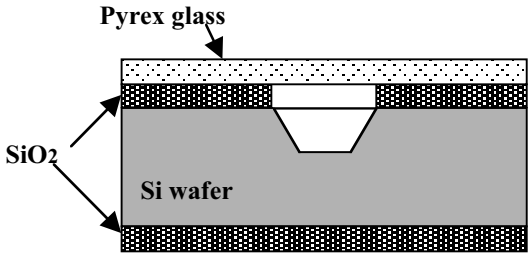

Figure 1: Cross section of the guide channel.

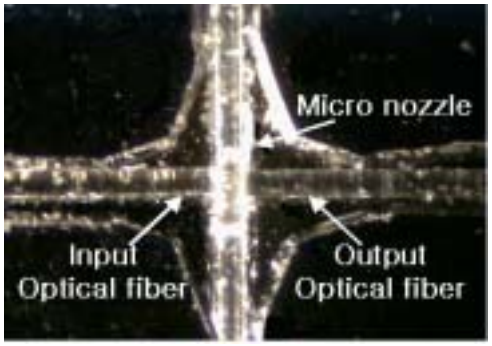

Figure 2: Photograph of fabricated guide channel.

micro nozzle and optical fibers, it was executed the anodic bonding(1000 VDC, 400 _) for pyrex glass and Si wafer which guide channel is fabricated on. The aligned photograph of optical fibers and micro nozzle on guide channel is shown in Figure 2.

In order to detect the small optical signal through the output optical fiber, we designed and made a signal processing circuit with photodiode(S2386-8K). We made the cell countable circuit with comparator(LM311). Figure 3 shows the block diagram of the fabricated circuits for an amplification and cell counting. We used data acquisition system(DaqBook/100) and personal computer to process the output signal from an electrical circuit. The digital oscilloscope(LeCroy LC534A) is used to measure effectively the changes of a light quantity. Then we made the program by Visual Basic 6.0 to observe the output of cell counter. Figure 4 shows the schematic diagrma of the total systems.

3 EXPERIMENT AND RESULT

In order to find the most suitable mode, we have measured the output light quantity of the output part with single mode and multi mode optical fiber. By injecting Huvec cells with phenol red stain in micro nozzle like Figure 5, we measured output power. The results are shown in Table 1. We used common optical fiber which its diameter is 125 _. We used a core size of single mode is

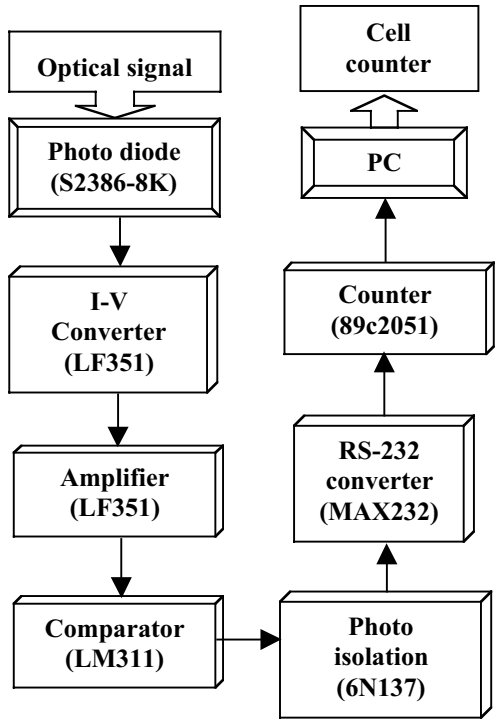

Figure 3: Block diagram of signal processing circuit.

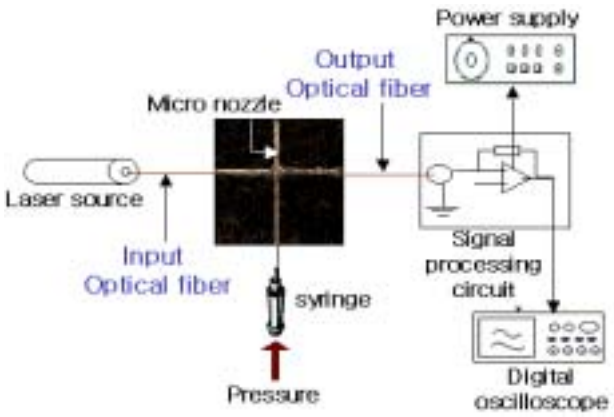

Figure 4: Schematic diagram of the proposed system.

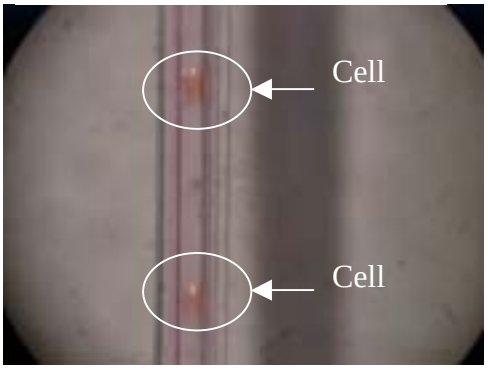

Figure 5: Photograph of flow cells in micro nozzle.

about 8.3 _ and it was difficult to align eventhough big power of light source. But the core size of multi mode is about 62.5 _ and it was easy to align. The output power of light through an optical fiber was big also.

Input fiber	Output fiber	Detection	Output power
single	single	impossible	
single	multi	possible	0.2_
multi	single	possible	0.6_
multi	multi	possible	0.12_

Table 1: Relation of between optical fibers and output power.

Therefore in case of single mode both input and output, we were not able to detect the flow cells. In the other hands, we were able to detect for a single mode in input and a multi mode in output. But it was difficult to align and the output voltage was low at about 80_. For multi mode both input and output, it was easy to align. The sensitivity of cell detection was not bad as other cases. Because the output voltage was high at about 300_, it is able to replace by a light source which its output power is small relatively. So it is able to use a laser diode and is suitable for a minimization. Figure 6 shows the voltage characteristic of Human T cell for single-multi modes. Figure 7 shows the result of Mouse T cell for multi-multi modes. The voltage characteristics is different because a light permeability is differnt for each cell which is made up of different cellulose and cell wall. In addition, we knew the relative cell size. Figure 8 represents the result of Mouse T cell for multi-multi modes. That is, Figure 6 and Figure 8 were measured at the same time under the same conditions. However, the pulse width of Figure 8 was one point five times as wide as the pulse width of Figure 6. That is to say, the cell size of Figure 8 was one point five times as big as the cell size of Figure 6.

In result of injecting Mouse T cells(EL-4) 6_10^5/_ in the fabricated system, it was able to detecte about 23000 cells for 8 minutes and 34 seconds. The average counting speed was 45 cells/sec. Figure 9 represents the result of fabricated cell counter.

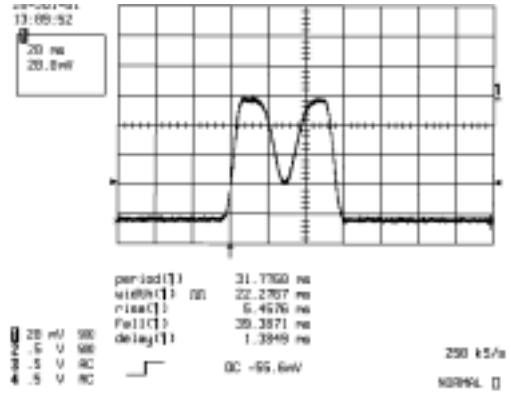

Figure 6: Voltage characteristic for Human T cell. (single-multi fibers, small)

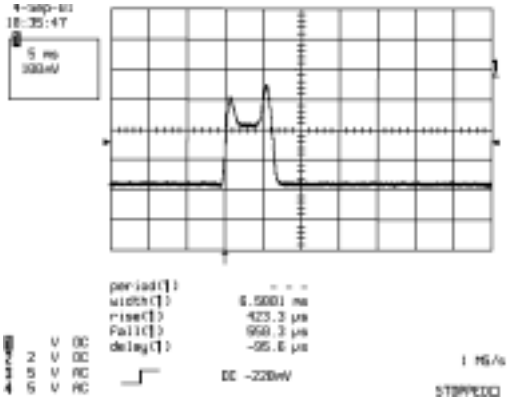

Figure 7: Voltage characteristic for Mouse T cell. (multi-multi fibers)

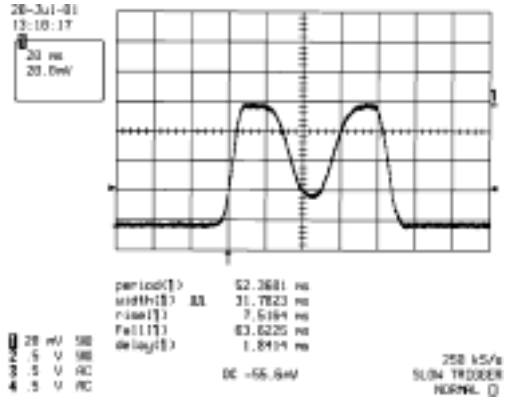

Figure 8: Voltage characteristic for Human T cell. (single-multi fibers, large)

Figure 9: Result of the fabricated cell counter.

4 CONCLUSION

In this experiment, we tested various optical fibers to select a suitable fiber for a effective flow cell detection. In order to align between micronozzle and optical fibers, a guide channel was fabricated by (100) silicon wafer etching with MEMS (Micro Electro-Mechanical System) technology. This fabricated system is advantageous due to its low cost and simple structure. This is possible because it replaces many optical lenses and expensive equipment with optical fibers. In result of cells detection by using various fiber modes in this system, for multi mode both input and output part, it was easy to align. The sensitivity of cell detection was not bad as other cases. The output voltage was as high as about 300_, so we are going to use a light source which has relatively small output power. Therefore, it was suitable for minimization. By injecting various cells, we were able to detect cells. The reason for the various voltage characteristics is the difference of light permeability for each cell which is made up of specific cellulose and cell walls. In addition, we were able to compare the relative size of the injected cells.

Therefore we show the capability of a minimization for common flow cytometry. This is able to realize a mass production of low cost by MEMS technology. In addition, we are able to apply this to the sorting system which sorts the specific cells among all kinds of cells. This system is able to apply to micro flow cytometry.

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REFERENCES

[1] Eric Altendorf, Diane Zebert, Mark Holl, and Paul Yager, "Differential Blood Cell Counts Obtained Using a Microchannel Based Flow Cytometry", Transducers 97, pp.531-533, 1997.

[2] L. Scott Cram, John C. Martin, John A. Steinkamp, Thomas M. Yoshida, Tudor N. Buican, Babetta L. Marrone, James H. Jett, Gary Salzman, and Larry Sklar, "New Flow Cytometric Capabilities at the National Flow Cytometry Resource," Proceeding of the IEEE, Vol. 80, No. 6, pp. 912-917, 1992.

[3] Howard M. Shapiro, M.D., "Practical Flow Cytometry", Third Edition, 1995.

[4] E. Kim, W. Kim, S. Park, K. Chun, J. Chang, S. Yang, J. Kim, "Development of Micro Flow Cytometric System", Proceedings of the ISC 2001, pp.159-160, 2001.

[5] L. Scott Cram, John C. Martin, John A. Steinkamp, Thomas M. Yoshida, Tudor N. Buican, Babetta L. Marrone, James H. Jett, Gary Salzman and Larry Sklar, "New Flow Cytometric Capabilities at the National Flow Cytometry Resource", Proceeding of the IEEE, Vol. 80, No. 6, pp. 912-917, 1992.

[6] David P. Schrum, Christopher T. Culbertson, Stephen C. Jacobson and J. Michael Ramsey, "Microchip Flow Cytometry Using Electrokinetic Focusing", Analytical Chemistry, Vol. 71, No. 19, pp. 4173-4177, 1999.

[7] Ye Yang, Zhen-Xi Zhang, Xin-Hui Yang and Da-Zong Jiang, "The Blood Cell Counting and Classification from Stationary Suspensions by Laser Light Scattering Method", Proceedings of the 20th Annual International Conference of the IEEE Engineering in Medicine and Biology Society, Vol. 20, No. 4, pp. 1885-1888, 1998.