

A Visual Feedback System for Micromanipulation with Stereoscopic Microscope

Tetsuya Sano

Faculty of Education Okayama University, Tsushima Okayama, 700, Japan
Phone: +81 (086) 251-7723, email: tsano@cc.okayama-u.ac.jp

Hidekazu Nagahata

Faculty of Education Okayama University, Tsushima Okayama, 700, Japan

Hiroshi Endo

School of Health Sciences Okayama University, Shikata Okayama, 700, Japan

Tetsuhiro Sumimoto

Maritime Safety Academy, Kure Hiroshima, 737, Japan

Takeshi Kunishi

Interface co.ltd, Minamiku Hiroshima, 732, Japan

and

Hideki Yamamoto

Faculty of Education Okayama University, Tsushima Okayama, 700, Japan

Abstract – A Stereoscopic microscope is widely used in a micromanipulation such as to operate genes and to inspect integration circuits. As in these tasks the micromanipulation is handled and makes too heavy burden to operators, it is desirable to perform the micromanipulation automatically. In this paper, we propose a visual feedback system for micromanipulation with stereoscopic microscope. This system takes less time to control the manipulator by reducing searching area to detect an object.

Keywords – Visual Feedback, Stereovision, Micromanipulation, Image processing, Stereoscopic Microscope

I. INTRODUCTION

A stereoscopic microscope is widely used in a micromanipulation such as to operate genes and to inspect integration circuits. As in these tasks the micromanipulation is manually handled and makes too heavy burden to operators, it is desirable to handle the micromanipulation automatically. In this paper, we propose a visual feedback system for micromanipulation with stereoscopic microscope.

The stereoscopic microscope observes an object three-dimensionally with stereovision. Two TV cameras is set to the eyepieces of the microscope. The distance between the object and object lens and the distance between the needle and the object lens are obtained by using stereovision method. The error signal based on the distance

between the object and the needle is derived. The micromanipulator is controlled to make this error signal to zero. The objects are colored beads with 2mm diameter instead of the cells. The needle 0.6mm in diameter 20mm in length is attached to the micromanipulator.

The detection of the corresponding point in the left and right images, measuring of the focal distance of the camera and the distance between the cameras become necessary in the stereo-vision method.

But, it is difficult to measure the focal distance and the distance of the cameras. Furthermore, it is difficult to secure the accuracy of distance measuring because a distance between the tip of the needle and the object is small in comparison with the distance between the object and the object lens. As the distance between the object and the needle is in proportion to the difference of view in the left and right images, this distance is calculated directly from the difference of view.

Corresponding point to the object is the centroid of the object, and it is the tip of the needle as for the needle. The object and the needle is detected by its color and shape.

The descent speed of the needle is small as the tip of the

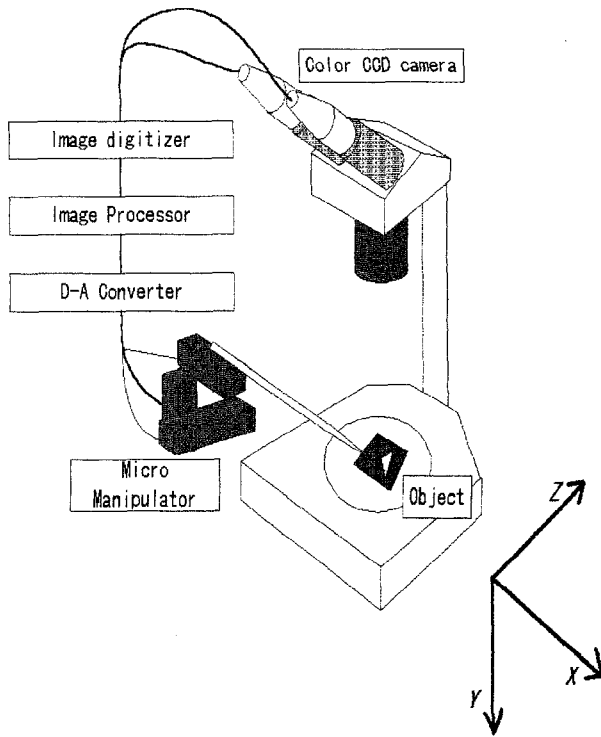


Fig. 1. Visual feedback system

needle approached the object to prevent an accident such as the destruction of the object by the needle piercing the object.

As the next position of the tip of the needle is predictable, the searching areas of the needle in left and right images can be reduced.

II. VISUAL FEEDBACK SYSTEM

Fig.1 shows the visual feedback system. This system is composed of two color TV cameras, a stereoscopic microscope, an image processing system and a micromanipulator. Two TV cameras are set to the eyepieces of the microscope and the left and right images are obtained from the TV cameras, respectively. An object is put on the stage of the stereoscopic microscope. A needle is attached to the micro-manipulator to the right in the left and right images. Lighting is given from the bottom of the stage, and the object is observed. A background is the lightest, and a needle is dark.

This visual feedback system controls the micromanipulator to reach the tip of the needle to the object. From the stereovision method, an error signal based on the distance between the object and the tip of the needle is derived.

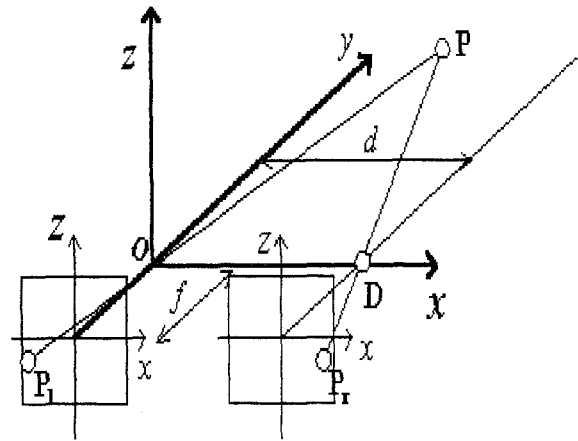


Fig. 2. Principle of stereo-vision method

The micromanipulator is controlled to make this error signal to be zero.

A distance between the object and the tip of the needle is expressed by X, Y and Z axis. As the control speed of the micromanipulator is fixed, its control time is estimated by the distance and the speed.

Two CCD cameras is set side by side. The horizontal position of the object and the tip of the needle is measured by the right camera, and the depth is measured by the difference between right and left images. Fig.2 shows the principle of stereo-vision method. Two TV cameras with focal distance f and with the distance between left and right camera d figures the position of the object $P\{x, y, z\}$ to $P_L\{x_l, z_l\}$ in left image and $P_R\{x_r, z_r\}$ in right image. The difference of view is appeared in the difference between x_l and x_r . The formula of the position P is given by

$$P = \left(\frac{x_l \cdot d}{x_l - x_r}, \frac{f \cdot d}{x_l - x_r}, \frac{z_l \cdot d}{x_l - x_r} \right) \quad (1)$$

As the CCD cameras measures the object through the stereoscopic microscope, it is difficult to estimate the focal distance and the distance between left and right cameras.

The distance between the tip of the needle and the object is small in comparison with the distance between the object and the object lens, since it is difficult to get the high accuracy of distance. The difference of view is almost in proportion to the those distance of the object in measure at short range.

A distance Y between the object and the object lens can

be written as

$$Y = A \times (x_l - x_r) + B \quad (2)$$

where x_l is the X coordinate of the object in left image, x_r is in right image and $(x_l - x_r)$ is the difference of view. A distance $(Y_2 - Y_1)$ between the object and the needle is given by

$$Y_2 - Y_1 = A \times (x_{l2} - x_{r2} - (x_{l1} - x_{r1})) \quad (3)$$

where the distance between the needle and the object lens is Y_1 , the distance between the object and the object lens is Y_2 , difference of view is $(x_{l1} - x_{r1})$, $(x_{l2} - x_{r2})$ respectively.

The movement of the needle on the image when a needle moves around the object is observed, a needle moves $Y_2 - Y_1$, and an image measurement does the movement quantity $x_{l2} - x_{l1}$ of the needle of the image and $x_{r2} - x_{r1}$. A is determined by the equation 3.

By using stereo-vision method, a distance between the object and the object lens is calculated by looking for corresponding point that the coordinate of the object is shown about left and right images. A corresponding point of the object is its centroid, and the point of the needle is the tip of the needle. The object and the needle is detected by its color and shape. The normalized area K is given by

$$K = \frac{L^2}{S} \quad (4)$$

where L is the length of the contour, and S is the size of the object.

The object in the images is detected from referring to the color. More than one error is detected. The object is detected from referring to the shape. But, a detected object often lacks the contour. Hence, the shape of the object changes, and specifying that object becomes difficult. Hence, the contour of the object is detected by using the laplacian filter. The shape of the object can be detected precisely as a result of making up the contour of the object.

III. ALGORITHM OF THE CONTROL OF MICROMANIPULATOR

The error of measurement of the distance has a danger of the needle piercing the object. As a system is initialized to measure a position in the neighborhood of the object, the accurate measurement of the distance between the object and the tip of the needle becomes difficult. When control is started, it is presumed a needle that it is away from the object. The manipulator is controlled to approach an object gradually with the repetition of measuring the distance.

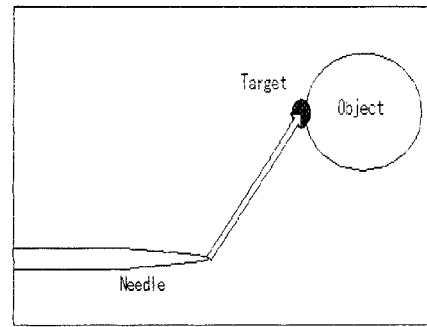


Fig. 3. The needle moves to the left of the object while the needle is at a higher level than the object.

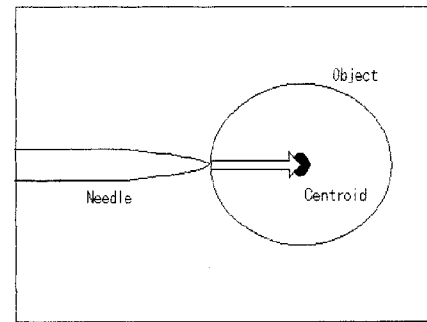


Fig. 4. The needle is made to reach it to the centroid after the needle is at the same level as the object.

The approach speed of the needle is restricted small as the needle approached an object to prevent from the needle piercing the object. And, detection at the needle becomes difficult when the needle comes near to the object because a color is almost the same in the contour of the object and the needle. While the needle is at a higher level than the object, the needle moves to the left of the object as shown in Fig.3. After the needle is at the same level as the object, a distance between the centroid of the object and the needle is measured, and the tip of the object is controlled to reach the centroid of the object as shown in Fig.4.

As a tip of the needle comes closer to the object, a measurement accuracy of the distance increases. Then the speed of the descending needle at the beginning of the control is faster than that at the end of the control.

The arrival distance is estimated about X, Y and Z axis direction from the precision of distance measuring. It considers that the needle reached it, and handling is finished when the distance between the object and the needle is smaller than the arrival distance.

The visual feedback system takes much time for distance measuring. There are many calculations along with the

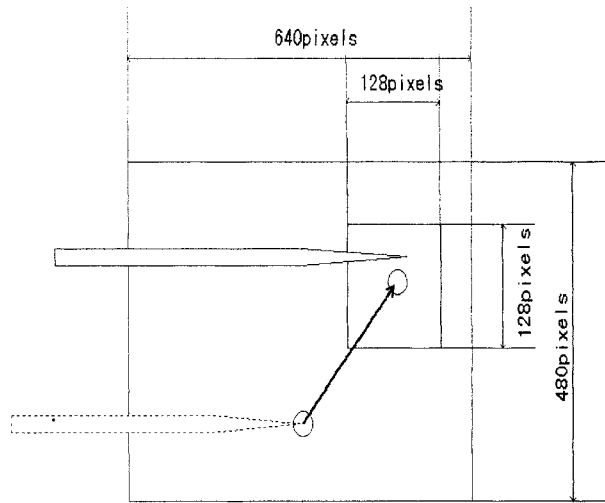


Fig. 5. Searching area

image analysis, and the calculating time occupies most of the control time of the system. As the next position of the tip of the needle can be predicted, the searching areas of the needle in left and right images become narrow. The amounts of calculation along with the image analysis are reduced, and handling is processed with high speed. The searching area is set up as shown in Fig.5.

The position of the tip of the needle on the image before control is found as P_0 . As the movements of the manipulator are Δx , Δy and Δz , its position after control is predicted as P_1 were the position that Δx and Δz were added to P_0 . A searching area is set up in the area of the 128×128 pixels around the point P_1 . The amount of calculation is decreased by set up the searching area of the needle small.

IV. EXPERIMENTAL RESULTS

The object is red bead with 2mm diameter, and a manipulator is controlled automatically to make the tip of the needle reach the centroid of the object. A ruler put on the stage of the micromanipulator is measured, and the length of 1 pixel is estimated. The position of the needle in the height which is the same as the object is found. The next position of the needle when it was raised 0.5mm from that position is found, and a coefficient A is estimated by the change of the difference of view. The movement length is shown in Table I. It is considered that the needle is reached when a distance between the needle and the object becomes within ± 0.27 mm about X, the Z axis direction about ± 0.21 mm, the Y axis direction. The movement distance when a manipulator is operated for 60 seconds in each of X, Y and the Z axis direction is

TABLE I
The movement length

X	3.519×10^{-2} mm/pixel
Y	1.667×10^{-1} mm/pixel
Z	3.464×10^{-2} mm/pixel

measured. The movement speed of the manipulator is as shown Table II. The control time of the manipulator is calculated referring to this table.

TABLE II
The movement speed of the manipulator

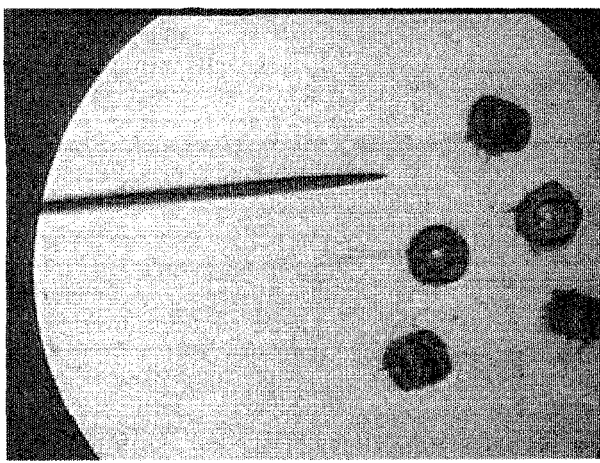
	+direction	-direction
X	3.783×10^{-2} mm/s	4.167×10^{-2} mm/s
Y	3.967×10^{-2} mm/s	3.500×10^{-2} mm/s
Z	4.267×10^{-2} mm/s	4.817×10^{-2} mm/s

Fig.6 shows the needle before control. The red bead which becomes an object is at the upper right of the image. The tip of the needle is almost located in the center of the image, for the right.

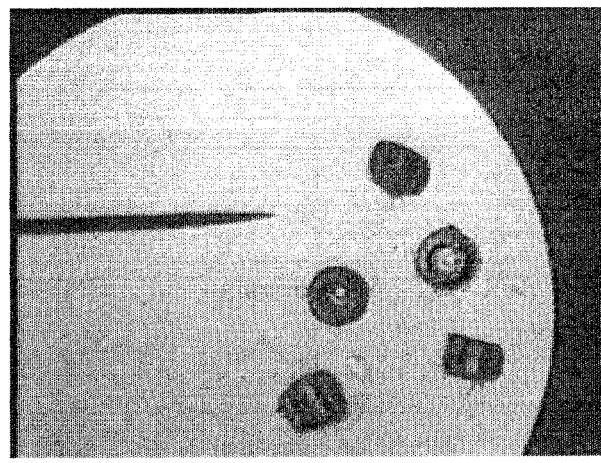
A region of interest is set up respectively about the needle and the object. A color is estimated from histograms of the region of interest. The shape of the object that is included in the region of interest is estimated.

Fig.7 shows the histogram of the region of interest include the object. An object appears as a dark distribution, and a background appears as the light distribution. A threshold is estimated from containing the distribution of showing an object. The object is detected in the image from referring to the threshold. Fig.8 shows the object detected in the images. The area of the object in the left image and the right image and a normalized area are looked for from each images. While a manipulator is controlled, the object and the needle are detected from the images referring to the color and the shape. The shape of the object of the left image and the right image is shown in Table III. It almost has a circle respectively, and a normalized area is small. The shape of the needle is shown in Table IV. A needle has slender shape, and the value of its normalized area is large.

The tip of the needle reached the centroid of the object as a result of doing three times in total, distance measuring and the control of the manipulator. Table V shows the distance between the object and the needle to the arrival from the start of the control.

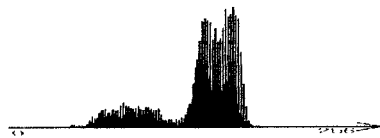


a) Left image

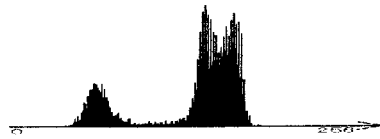


b) Right image

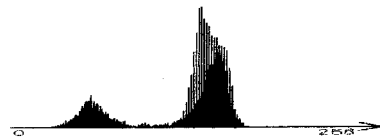
Fig. 6. The needle under moving



a) Red color component



b) Green color component

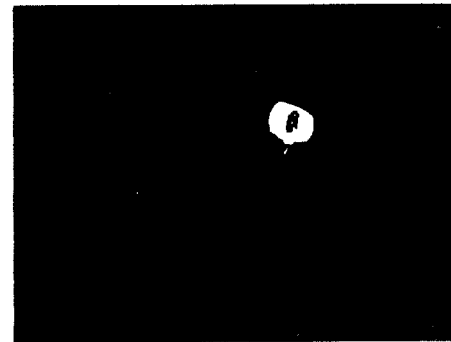


c) Blue color component

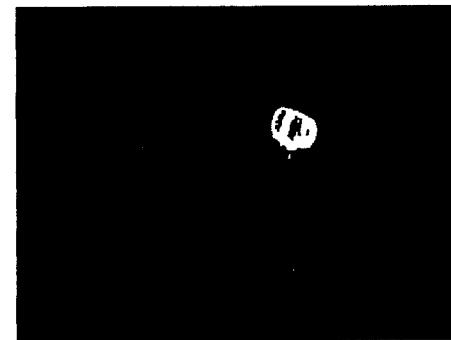
Fig. 7. The histograms of the ROI including the object (Red bead)

The tip of the needle reached above the left of the object by the first control. The tip of the needle reached the left of the object by the second control. An object is approached, and a focus is decided with the needle. An image becomes vivid, and the precision of distance measuring improves. A distance between the object and the tip of the needle was measured in the third time, and a needle was handled to reach an object.

Fig.9 shows the needle reached the object. The tip of the needle reaches the neighborhood of the top of the object, the centroid.



a) Left image

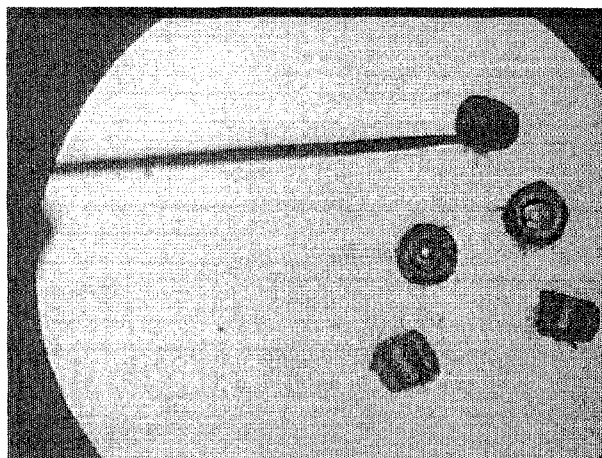


b) Right image

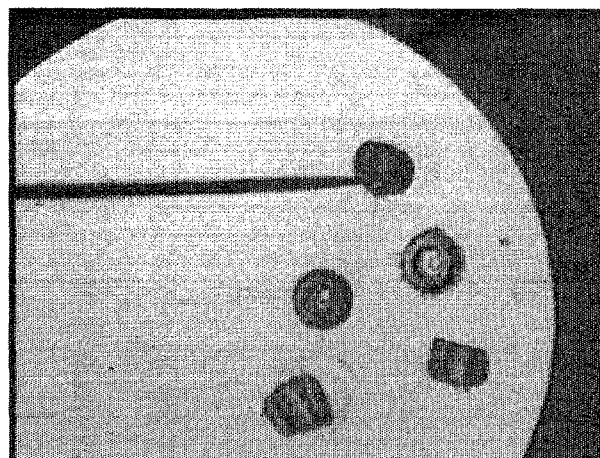
Fig. 8. Detected object.

TABLE III
The shape of the object

	Left	Right
Size (pixels)	3246	2433
Normalized Area	17.88	20.84



a) Left image



b) Right image

Fig. 9. The needle reached the object

TABLE IV
The shape of the needle

	Left	Right
Size (pixels)	1124	1111
Normalized Area	42.9	56.6

TABLE V
A distance between the needle and the object

	X	Y	Z	To
1	3.201	0.333	-1.282	The left of the object
2	0.000	0.083	0.000	The left of the object
3	2.111	0.833	0.000	The centroid

V. CONCLUSIONS

The visual feedback system which automates the work, which handles a tip of the needle reach an object under the stereoscopic microscope was developed. An object and a needle are measured by using two cameras.

A needle goes by mistaking distance measuring to the object from the needle, and the accident which destroys the object happens. A manipulator was controlled so that an object might not pile up with the needle on the image not to do wrong distance measuring.

Because the position of the next needle in image can be predicted, the searching area to detect the needle is narrowed, and the amount of calculation along with the image analysis is reduced. Consequently, the system takes less time to detect the needle. Hence, this proposed system can be adapted in the micromanipulation with the stereoscopic microscope.

References

- [1] Huang, T.S, Image sequence analysis, Springer-Verlag, 1981.
- [2] Zhang, Z and Faucher, O, 3D dynamic scene analysis, Springer-Verlag, 1992.
- [3] Sankar, K.P, Dwijesh, K., and Dutta, M, Fuzzy Mathematical approach to pattern recognition, John Wiley & Sons, 1986.
- [4] Liu, J. and Yang, Y., Multiresolution color image segmentation, IEEE Transactions on Pattern Analysis and Machine Intelligence, Vol.16, No.7(1994), 689-700.
- [5] Castano, A and Hutchinson, S, Visual compliance: Task-directed visual servo control, IEEE Transactions on Robotics and Automation, Vol.10, No.3(1994), 334-341.
- [6] Volpe, R and Khosla, P, Analysis and experimental verification of a fourth order plant model for manipulator force control, IEEE Robotics & Automation Magazine(1994), 4-13 Vol.10, No.3(1994), 334-341.