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# **Thickness and Refractive Index Measurement System for Multilayered Samples**

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**ABSTRACT** In order to simultaneously obtain the thickness and refractive index for each layer of multilayered samples, this paper proposes a novel measurement system with a simple structure by using geometric optics. The key point of the overall structure is that the binary linear equation relation between the upper and lower planes of the layer can be known by the distances between laser spots reflected from the layer. Using two optical paths with different incident angles of laser beams, the CCD sensors capture the spot signal for image processing binarization and data sampling. After the obtained laser spots' spacing is substituted into the equations, the thicknesses and refractive indexes of the multilayered samples can be calculated and measured. The proposed measurement system is characterized numerically using simulations on the commercial software program Zemax and then experimentally tested using a laboratory-built prototype. The experiment results show that the refractive indexes and the thicknesses of three-layer samples were measured with high accuracy (with maximum measurement errors of 2.4% and 2% for a refractive index n and thickness t, respectively).

**INDEX TERMS** Auto-focusing microscopy, multilayered samples, optical system, refractive index, thickness.

#### I. INTRODUCTION

For a long time, optical technology has been an important tool for non-destructive testing [1]. Especially in recent years, the precision miniaturization of electronic products has become a mainstream trend. Production and testing equipment for component processing and assembly has also received more attention [2], [3]. How to ensure product quality control is a very important issue. In manufacturing and inspection fields, machine vision systems are increasingly used due to their reliability, relatively low cost and high throughput. It is usually necessary to focus the camera to obtain a clear image of the target object [4]–[13]. However, for the existing optics-based microscopes, they are only suitable for single layer of samples or reflected surfaces [14]-[21]. For the biomedical inspection, three-layer samples (for example, the cover glass, biological sample, and slide glass) are popular. Consequently, manual focusing microscopes are widely used and they are time consuming. Therefore, it is a critical issue to simultaneously obtain the thickness for each layer of multilayered samples to implement the auto-focusing function.

In addition, biological testing is also in greater demand due to the development of minimally invasive surgery. The development of laser therapy and other precision surgery has higher requirements for understanding the phenomenon of light transmission and refraction between biological tissues. Especially when using an optical coherence tomography system, it will affect the contrast of the image and the subsequent calculation of tissue thickness [22]–[24]. As a result, there are various measurement techniques proposed over the last decade for the simultaneous measurement of the refractive index and thickness of samples [25]–[44]. However, to the best of our knowledge, there is not any existing method to simultaneously measure the thickness and refractive index for each layer of multilayered samples.

Therefore, in this paper, the proposed measurement system was able to simultaneously obtain the thickness and refractive index for each layer of multilayered samples. The proposed measurement system was characterized numerically, after which the results were verified experimentally using a laboratory-built prototype.

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FIGURE 1. Structure of proposed measurement system.

### II. PROPOSED MEASUREMENT SYSTEM WITH DIFFERENT INCIDENT ANGLES

#### A. STRUCTURE

The structural design of the proposed measurement system is inspired by the triangulation distance measurement. As shown in Fig. 1, two sets of reflective geometric optical architectures with different incident angles are designed for measurement. The two sets of mathematical formulas from the two different incident angles can mutually calculate the respective thickness and refractive index for each layer of multilayered samples. The proposed measurement system can be integrated into an auto-focusing system for further auto-focusing applications. In the proposed measurement system, the laser on the main optical axis is first split into two optical paths through a beam splitter (BS), and the laser beam in each optical path passes through a convex lens (CL<sub>1</sub>, CL<sub>2</sub>) for laser beam shaping, respectively. In order to produce two different incident angles  $(30^{\circ} \text{ and } 45^{\circ})$ , the laser beams in two optical paths will pass through a mirror  $(M_1, M_2)$ , respectively. The angles of 30° and 45° are chosen for the biomedical inspection of three-layer samples (thickness of 0.3 mm  $\sim$  2 mm) with suitable measuring accuracy. The laser beams reflected from the upper and lower surfaces of the samples will be focused on two charge-coupled devices (CCD<sub>1</sub> and CCD<sub>2</sub>). Consequently, MATLAB software was used to perform the proposed digital image processing technique to simultaneously obtain the thickness and refractive index for each layer of multilayered samples. By using the thicknesses obtained by the proposed measurement system, the motor of the auto-focusing system can be controlled to move the objective lens to achieve the auto-focusing function on the target surface.



FIGURE 2. Schematic illustration of optical path in proposed measurement system.

#### **B. MEASUREMENT PRINCIPLE**

The aim of this paper is to measure the physical thickness and refractive index for each layer of multilayered samples, in special, for biomedical applications. In this paper, three-layer glasses were used to represent the cover glass, biological sample, and slide glass. When stacking three pieces of glasses, there are two air gaps between glasses. Therefore, the reflected laser spots are 6 points. The basic principle of the mathematical model proposed in this paper is shown in Fig. 2. As laser beam enters the glass sheet, it will produce both refraction and reflection. According to the principle of geometric optics, the relationship between the light spots on the CCD sensors and the thicknesses and refractive indexes of the three-layer samples can be calculated.

Since six unknowns of thicknesses and refractive indexes are required, this issue is solved with two different incident



FIGURE 3. Detailed illustration of different incident angles.

angle beams, as shown in Fig. 3. According to the geometric relationship described in the figure and Snell's Law, we can get the relationship between the spot distance (r) on the CCD sensor and the refractive index (n) and thickness (t) of the sample:

$$\begin{bmatrix} r_{a1} = 2 \cdot t_1 \cdot \tan \left[ \sin^{-1} \left( \frac{\sin \theta_a}{n_1} \right) \right] \\ r_{a2} = 2 \cdot t_2 \cdot \tan \left[ \sin^{-1} \left( \frac{\sin \theta_a}{n_2} \right) \right] \\ r_{a3} = 2 \cdot t_3 \cdot \tan \left[ \sin^{-1} \left( \frac{\sin \theta_a}{n_3} \right) \right] \end{bmatrix}$$
(1)

By dividing (1) and (2), we can get:

$$\frac{r_{a1}}{r_{b1}} = \frac{\tan\left[\sin^{-1}\left(\frac{\sin\theta_a}{n_1}\right)\right]}{\tan\left[\sin^{-1}\left(\frac{\sin\theta_b}{n_1}\right)\right]} \tag{3}$$

$$\frac{r_{a2}}{r_{a}} = \frac{\tan\left[\sin^{-1}\left(\frac{\sin\theta_{a}}{n_{2}}\right)\right]}{\left[1 + 1 + 1\right]} \tag{4}$$

$$\frac{r_{b2}}{r_{a3}} = \frac{\tan\left[\sin^{-1}\left(\frac{\sin\theta_b}{n_2}\right)\right]}{\left[\frac{r_{a3}}{n_2}\right]}$$
(5)

$$\frac{ds}{r_{b3}} = \frac{1}{\tan\left[\sin^{-1}\left(\frac{\sin\theta_b}{n_3}\right)\right]}$$
(5)

In (3), (4) and (5), the distances and angles are known, so the refractive index of each layer can be obtained. Then the thickness can be obtained by substituting the refractive index of each layer into (1) and (2).

#### III. OPTICAL SIMULATION OF PROPOSED MEASUREMENT SYSTEM AND IMAGE PROCESSING PROCEDURE

In this section, a series of numerical simulations using the software program Zemax were performed to design and verify the measurement performance of the proposed measurement system. In the proposed measurement system, the convex lenses ( $CL_1$ ,  $CL_2$ ) are designed to shape the laser beam spot, so Zemax simulations can be used to confirm the shaping effect.



FIGURE 4. Zemax 3D optical model of proposed measurement system.



FIGURE 5. Zemax 3D optical model of proposed measurement system.

Figure 4 shows the Zemax 3D optical model of the proposed measurement system. Figures 5a and 5b show the simulated laser spots on  $CCD_1$  without and with the convex lens, respectively. It can be clearly seen that the plastic effect obviously separates the laser spots, which improves the post-image processing efficiency. The selected design parameters of the proposed measurement system are listed in Table 1.

To determine the spot distance (r), we used the proposed digital image processing technique and MATLAB software to process and analyze images captured by the CCD sensors. The proposed digital image processing technique involves three fundamental steps: (1) image median filtering, (2) image binarization, and (3) data sampling. Figure 6 shows the original laser spot image made by Zemax; the image processing results aid in retrieving the distances between laser spots. As shown, the distances between the laser spots in the images captured by the CCDs can be calculated. A more comprehensive description of the image processing technique is provided in our previous studies [20], [45], [46].

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FIGURE 6. Image processing procedure and results.

TABLE 1. Design parameters of proposed measurement system.

Component	Brand	Specification
Laser	Thorlabs	HL6501MG
BS	Thorlabs	CM1-BS013(50:50)
$M_1, M_2, M_3$	Thorlabs	PF10-03-P01-10
$CL_1$	Thorlabs	LA1484, $f_0=300 \text{ mm}$
$CL_2$		LA1172, $f_0$ =400 mm
		Sapphire Window
Sample	Edmund	ZnSe Window
		Magnesium Fluoride Window
Objective Lens	Olympus	$f_0=18 \text{ mm}$
$CCD_1, CCD_2$	Basler	5472*3648 (pixel), 17fps
$CCD_3$	Duma	1280*1024 (pixel), 50 fps
Infinity-Corrected	Novito	1 60255
Optics System	Inavita	1-00255
Motor	PI	UPL120, 13 mm
meter	11	01 £120, 15 mm

To verify that the proposed measurement system can simultaneously measure the thicknesses and refractive indexes of three-layer samples, four common transparent materials such as sapphire, borosilicate glass (BK7), magnesium fluoride glass (MgF2), and zinc selenide glass (ZnSe) were simulated. The thickness of BK7 that represent microslide with a refractive index of 1.5168 is 0.7 mm; the thickness of MgF2 with a refractive index of 1.3777 is 2 mm; the thicknesses of sapphire with a refractive index of 1.7682 are 1mm and 2mm, respectively; and the thickness of ZnSe with a refractive index of 2.3674 is 2 mm.

Figures 7 and 8 present two sets of simulation results for the simulated image and the calculated thicknesses and refractive indexes, respectively. The first set of samples are with the upper 2 mm MgF2, the middle 1 mm sapphire, and the lower 0.7 mm BK7. The second set of samples are with the upper layer 0.7 mm BK7, the middle layer 2 mm sapphire, and the lower layer 2 mm ZnSe.

From the simulation results, the calculated thickness error and refractive index error are about 0.05% and 0.1%,



FIGURE 7. Simulation results for first set of samples.



FIGURE 8. Simulation results for second set of samples.



FIGURE 9. Laboratory-built prototype of proposed measurement system and sample.

respectively. Due to its low deviation value and high accuracy, the feasibility of this mathematical model and the proposed measurement system can be proved.



FIGURE 10. Experimental results for first set of samples.



FIGURE 11. Experimental results for second set of samples.

#### **IV. EXPERIMENTAL RESULTS AND DISCUSSION**

The validity of the proposed measurement system was verified using a laboratory-built prototype. Figure 9 shows a photograph of the laboratory-built prototype and sample.

In this section, a total of 5 glass sheets were measured, including 4 kinds of materials, and the true thicknesses were obtained through a commercial coordinate measuring machine (CMM, ZIESS Calypso). The thickness of MgF2 with a refractive index of 1.377 is 2.079 mm; the thicknesses of sapphire with a refractive index of 1.77 are 1.029 mm and 2.052 mm, respectively; the thickness of ZnSe with a refractive index of 2.631 is 2.058 mm; and the thicknesses of a general glass slide of unknown material is 0.707 mm. Take three of these arrangements and measure the thicknesses and refractive indexes of three glass slides simultaneously. Figures 10 to 13 show the experimental images and measurement results of 4 sets of samples, respectively. The first set



FIGURE 12. Experimental results for third set of samples.



FIGURE 13. Experimental results for fourth set of samples.

of samples are with the upper layer 2 mm MgF<sub>2</sub>, the middle layer 1 mm sapphire, and the lower layer 0.7 mm microslide. The second set of samples are with the upper layer 2 mm sapphire, the middle layer 2 mm MgF<sub>2</sub>, and the lower layer 2 mm ZnSe. The third set of samples are with the upper layer 2 mm sapphire, the middle layer 1 mm sapphire, and the lower layer 0.7 mm microslide. The fourth set of samples are with the upper layer 2 mm ZnSe, the middle layer 0.7 mm microslide, and the lower layer 1 mm sapphire.

From the experimental results, it can be deduced that the measured errors for thickness and refractive index in the actual experiments are about 2% and 2.4%, respectively. The errors for thickness are gotten from disparity between the measured thickness with the proposed measurement system and the measured thickness with the CMM. The errors for refractive index are gotten from disparity between the measured refractive index with the proposed measurement system



FIGURE 14. Laboratory-built prototype of auto-focusing system.



FIGURE 15. Focus image.

and the standard value with samples. The measured errors are obviously greater that the calculated errors. This is because the surface curvature (upper and lower) of each uniform refraction index layer, non-parallelism of the same optical borders in practice, and structural impurity of the layers material etc. will seriously transform the canonic application of Snell's law. As a result, the laser spots on CCD sensors are not in alignment well, as shown in Figs. 10 to 13 when comparing Figs. 7 and 8. In the future work, the non-parallelism of the samples should be considered into the proposed measurement system to improve the measuring accuracy.

Finally, the proposed measurement system was integrated into an auto-focusing system to achieve the auto-focusing function on the target surface with the three-layer samples of the upper layer 2 mm sapphire, and the middle and lower layers 0.7 mm microslides, as shown in Fig. 14. The focus

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target sample between each piece is scraps of lens cleaning paper. Figure 15 shows its auto-focusing image. The experimental results show that the proposed measurement system can successfully be integrated into an auto-focusing system for further biomedical application in the future.

#### V. CONCLUSION

This paper currently proposes an innovative, simple, and low-cost architecture for the future development of biological auto-focusing technology. It can simultaneously measure the thickness and refractive index for each layer of multilayered samples before focusing to achieve the purpose of rapid autofocusing. The experimental results show that measured errors for thickness and refractive index are about 2% and 2.4%, respectively. Consequently, the proposed measurement system has the potential to be used in the biomedical automated inspection.

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