Development of light scattering methods for urinary stones diagnosis

Y Warty, I A Fitri, F Haryanto and Herman
Institut Teknologi Bandung Jl. Ganesha 10 Bandung 40132, Indonesia
Email: yuniwarty@s.itb.ac.id

Abstract. The Kidney stones in the urinary tract area formed due to many factors. Most medical cases, this disease was diagnosed after the order of millimeters or centimeters. The purpose of this study was to characterize standard polystyrene solutions with light scattering techniques as a calibration tool that will be used to characterize urine crystals. The first stage begins with a literature review and optical settings. The second stage, the arrangement, and components of the tool were validated by measuring the aperture width based on the experiment and measuring the most appropriate cuvette to be used in the sample. The third stage, polystyrene was dissolved with 10 ml of tetrahydrofuran and measured by variations in molecular weight and concentration of polystyrene. The molecular weight of polystyrene was 4000, 90000, 200000, 400000 and 900000 grams/mol. The results showed the width of the gap according to the experiment was 0.05 mm. The cuvette which has a diffraction width value similar to the measurement without cuvette (Blank) measured from the central beam axis to dark 1 was a cuvette diameter of 27 mm. The results of the polystyrene solution showed a diffraction width change in the polystyrene solution for each molecular weight and concentration change. The higher the molecular weight and concentration, the smaller the diffraction width.

1. Introduction
Kidney stones are diseases of the urinary tract that occur due to the formation of hard material that resembles stones in the kidneys and urinary tract. This disease was recognized since ancient Greece [1]. The percentage of patients in each country varies like 1-5% in Asia, 5-9% in Europe, 13% in North America, 20% in Saudi Arabia [2]. This is also the third largest disease case in the urology department worldwide [3]. The high number of kidney stone sufferers also occurs in Indonesia. For example, the Cipto Mangunkusumo Hospital in Jakarta handles cases of kidney stones 5,174 / 6 years with a mean size of the stone length of 11.90 ± 7.54 mm [4].

Early formation of kidney stones due to the presence of supersaturation in the process of urine formation [5,6]. At this condition, the nucleus will be form and certain time interval form an aggregate. Kidney stones will develop to millimeters or even centimeters [7,8]. Patients usually feel pain after the size of the stone several mm also though the smaller size should be detected by urine. Several previous studies reported that several types of crystals can be observed in human urine such as oxalate, phosphate, uric acid [9]. Analysis of urine deposits also found in magnesium ammonium phosphate, apatite carbonate and mono ammonium vein [10].

The method that used as early detection of kidney stones was needed. Light scattering method is one method that can be used to diagnose the presence of crystals forming kidney stones in urine. This method was used before and showed the autocorrelation time of urine that comes from healthy people for 5
seconds. This time dropped 2-3 seconds by the presence of albumin, red blood cells, white blood cells, epithelial cells in the urine, and increases when there was oxalate [11]. Besides, the particle size distribution of urine in healthy people with laser scattering spectroscopy was 20 to 250 nm and compared with urine from unhealthy people 50-700 nm [12].

In this study, a light scattering method was developed to diagnose the presence of kidney stone crystals in the urine. This study was developed more comprehensively so that it can be utilized by the wider community as early detection of kidney stones. As a first step in this study, we calibrated the tool. So the specific purpose in this paper is to calibrate optical sets based on light scattering methods using a simulated solution (polystyrene) which will be a database for analysis of crystalline urine particles.

2. Methods

2.1. Material
Polystyrene (PS) (C8H8)n was used as a sample in this study. This material has a density of 0.96-1.04 g/cm³ and soluble in acetone, toluene, and tetrahydrofuran. The PS sample used varied molecular weight (4000, 90000, 200000, 400000, 900000) g/mol. The solvent used in this study was tetrahydrofuran because it has the highest solubility value compared to other solvents which have 0.96 g/mol. Sample preparation begins with weighing PS at each molecular weight of 2.6 mg, 5.2 mg, 7.8 mg, 10.8 mg 13.0 mg and mixed with 10 ml tetrahydrofuran. In addition, measurements were also made by dissolving PS on each molecular weight of 3.0 mg with 10 ml of tetrahydrofuran.

2.2. Optical alignment and baseline measurement
Validation of tool settings by measuring the slit width used based on experiment and comparing with the width of the gap written in the tool (0.05 mm). Variations in the distance between the gap with the detector (17 cm, 22 cm, 33 cm, and 14.6 cm) were intended to ensure the width of the gap in the tool. In addition, round cuvette with variations in thickness and diameter was measured to determine the most suitable cuvette for this study. The cuvette diameter was 16, 21, 23, 27, 30 mm.

The components of the tool in this study were laser light sources, a set of optical elements, samples, detectors, and computers for experimental control. The arrangement of tools as shown in Figure 1.

![Figure 1. The arrangement of the optical device settings](image)

Laser with a wavelength of 623 nm and a power of 1 mW has a high intensity which affects the intensity measured at the detector. The detector saturates because it was only able to measure the highest intensity at 65536. Therefore, a pinhole was used to reduce the intensity of light entering the detector. Concave and convex lenses applied to spread and collect signals. Then, the aperture to reduce the intensity that passes through the sample. The detector as a signal collector was Alphalas CCD-S3600-D (-UV) high-sensitivity linear CCD array with 3648 pixels, 16-bit ADC, 32MB RAM and USB 2.0. The detector has photoactive which is a linear array of individual pixels where 1 pixel is equivalent to 8 µm.
These pixels are sensitive to light and function to collect electric charges. The charge is converted to the value of digital light intensity by an analog-to-digital converter (ADC). The data obtained was stored in RAM and sent to the PC via USB. The signal was analyzed by calculating the diffraction width of the central beam axis to point 1.

3. Results and discussion

3.1. Optical alignment and baseline measurement
The width of the gap based on the experimental results with variations in the distance of the aperture to the detector was 0.05 mm. This value was the same as the number shown at an aperture of 0.05 mm. Measurements on several cuvettes produce different diffraction widths as shown in figure 2 (a) (b). This difference was caused by differences in cuvette diameter and cuvette thickness used. Two cuvettes with the same thickness were considered for use in the next study, cuvette 27 mm and 16 mm in diameter. Based on the diffraction width of the two cuvettes, the diffraction width value closest to the diffraction width without cuvette (blank) was the cuvette diameter of 27 mm shown in figure 2 (b).

![Figure 2](image.png)

**Figure 2.** (a) The diffraction pattern of various cuvette diameter (b) A graph on the relationship between the distance of the central beam axis - first diffraction minimum to cuvette diameter

3.2. Tools Calibration
PS dissolved in Tetrahydrofuran was measured for each molecular weight with concentration variations. The resulting signal pattern is in the form of diffraction as shown in figure 3.
Figure 3. The diffraction patterns of various polystyrene concentration

Figure 4. (a) A graph on the relationship between the distance of the central beam axis - first diffraction minimum to the addition of PS solution. (b) A chart on the relationship between the distance of central beam axis - first diffraction minimum to molecular weight

The difference in diffraction width at each molecular weight and the difference in concentration calculated from the central beam axis to the very point is shown in figure 4 (a). From the picture, it was observed that the greater the molecular weight, the smaller the width of the diffraction. The same thing was also observed in concentration variations where the graph results showed that the higher the
concentration of polystyrene the smaller the diffraction width. Besides that, at high molecular weights, the addition of polystyrene concentrations above 5.2 mg has little effect on diffraction width. This was shown in the graph of polystyrene with a molecular weight of 900000 g/mol with similarities in diffraction width for the addition of 7.8, 10.4, 13.0 mg PS.

Changes in the diffraction width of the central beam axis to the observation point at different molecular weights are shown in figure 4 (b). Each molecular weight of 3.0 mg was dissolved with 10 ml of Tetrahydrofuran showing different diffraction widths. The greater the molecular weight, the smaller the diffraction width formed. The molecular weight of 900000 g/mol means that the number of atomic weights that make it up was quite high so that it can be interpreted as polystyrene radius in solutions with a higher molecular weight greater than the lower molecular weight. These results will be used to observe kidney stone particles in the urine and become our concentration in the next study. In addition to this, the analysis of signals with Fourier transformation and calibration with standard tools will be applied.

4. Conclusions

The result of this research showed that the larger molecular weight reduces not only the intensity value but also the value of the width of diffraction. Further analysis of polystyrene is required. This analysis used as a reference for the measurement of particles in the urine sample.

References

[1] Modlin M 1980 S Adr Medical Journal 58 652-655
[2] Jing Z, Guozeng W, Ning J 2010 38 111-115
[3] Selvaraju R, Thiruppathi G, Raja A 2012 Molecular and Biomolecular Spectroscopy 93 260-265
[4] Noviandri E, Birowo P, Rasyid N 2014 Med J Indonesia 24 234–238
[5] Fukushima H, Ichiyanagi O, Kakizaki 2016 Urolithiasis 44 529-539
[6] Shamena A A, Arul T, Kumar R S, Kaltura S N 2015 Molecular and Biomolecular Spectroscopy 134 442-448
[7] He J Y, Deng S P, Ouyang J M 2010 Transacon nano bioscience 9 156-163
[8] Khan S R, D J K 2004 Pubmed 9 1450-1482
[9] Menon M, Parulka B G, Drach G W 1998 Campbell's urology 2661–2733
[10] Miano R, Germani S, Vespasiani G 2007 Urol Int 79 32–36
[11] Chiceaa, D., Chiceaa, I. M. 2007 Journal of Optoelectronics and Advanced Materials 9 694-697

Acknowledgments

The authors would like to thanks to Asahi Glass Foundation and Institute for Research and Community Services (LPPM), Institut Teknologi Bandung (424/I1.COI.PL/2017)