

CHAPTER V

CONCLUSIONS

In recent years, digital image-based colorimetry (DIC) has become more interested for color detection. There are several reports that reviewed the DIC in analytical chemistry. However, there was no report on use of CMOS webcam camera combination with artificial neural networks (ANNs). Therefore, the DIC-ANNs was selected in this study. In addition, the proposed method was applied for the determination of the extractable protein in natural rubber (NR) latex and medical latex gloves.

Effect of background on the color of the protein solution was studied in order to give reproducible color measurements. The designed sample cell holder building by using white PTFE gave a true color image of the solution. The reflected light could be reduced by using the sample cell holder for photography. Therefore, the sample cell holder made from PTFE was chosen as the optimum background for the other optimization experiment.

Effect of the focusing distance from camera lens to sample cell was studied by using automatic focusing of the CMOS webcam camera. The image obtained from the focusing distance at 7 cm showed the size of the sample cell bigger than those obtained from the other lengths. This provided the whole image which was easy to crop for image processing. Therefore, the focusing distance at 7 cm was selected for the further investigation.

Effect of the position of high intensity LEDs (one and two LEDs) on color of an image was studied. The best image was obtained by using two LEDs setting as shown in experiment set up 6 (Figure 39) which gave true color of solution in the sample cell holder and the sample cell. Nonetheless, the light of LEDs are straight line that cause shadow at the edge of the sample cell. Therefore, the angle of two LEDs with respect to the sample cell surface was studied at the angle of 30, 45, 60, and 90 degree in order to get the homogeneous image. It was observed that the most homogeneous image was obtained when setting the angle at 45 degree. Therefore, the

angle of the two LEDs setting at 45 degree with respect to a sample cell surface was chosen for the next optimization stage.

Effect of the light illumination of the LEDs was examined between 0-700 lux. The results observed that RGB values increased with increasing the light illumination up to 200 lux after which the RGB values became constant which could be attributed to the low light intensity that is not sufficient for the camera aperture. When increasing the light illumination until at sufficient or more intensities for the camera aperture, the light was controlled by automatic adjusting of the camera aperture causing the true color image which gave the constant RGB values. Therefore, the light illumination at 300 lux was selected for further image capturing.

The prediction performance of the written program was validated by the means squared error (MSE) and the relative standard error (%RSE). It was found that the average MSE of method 1 was lower than method 2 and 3. The lowest MSE indicated that accuracy of method 1 was higher than that of the other methods. Method 1 used the average of RGB-based value that was in wider range (0-16777215) than the other methods causing less error values when processing and providing low MSE. As compared to method 2 and 3 which used the average and mode number of R, G and B value (0-255), when these numbers were converted to RGB-based values, the high difference value attributed to high MSE was obtained. Thus, method 1 was chosen for the prediction of the protein in NR latex and medical latex gloves. The %RSE obtained was 0.05 when reading the RGB values for 3000 times. This indicated that the proposed method gave high precision. Therefore, the numbers of iteration for 3000 times were selected for the RGB values reading in the written program. The minimum and maximum levels of protein determination by proposed method were 1 and 100 $\mu\text{g mL}^{-1}$, respectively.

The determination of aqueous extractable protein in NR latex and medical latex gloves after color forming by the modified Lowry method by using the DIC-ANNs and spectrophotometry gave no statistical difference ($p>0.05$). The effect of substances that interferes with Lowry protein assay, including lipids, nucleic acids, and sugars were not study in this research because these could be eliminated by acid precipitation step before color developing [2, 92]. The amounts of the protein in NR latex by using the DIC-ANNs and spectrophotometry was 50.5(\pm 1.81) and 62.3(\pm 2.29)

$\mu\text{g g}^{-1}$, respectively. The concentration of the protein observed in NR latex samples can cause allergic to sensitized user [19]. The amount of the protein in medical latex gloves samples observed in Saf-gard is $433.1(\pm 15.0) \mu\text{g/glove}$ which was more than other brands. Nevertheless, all latex gloves samples have high protein content because they are powdered gloves which soluble protein can absorb in starch during processing [19]. The proteins in medical latex gloves samples can cause allergic to users who are sensitized when contact with skin or inhalant the starch on the latex gloves samples. Therefore, glove manufacturing which produces powdered gloves with reducing protein content which greatly reduces the allergenic potential is important to users [1].

Suggestions for future works

The written software can be improved for further works so that many processes can be automatically done. The DIC lightbox could be developed to another works such as color developing on plate or test kits. New software may be developed to show and process the consistent value in each measurement. More research work could be acquired for applying the DIC-ANNs in another colorimetric reaction.

It could be summarized that the advantages and disadvantages of the proposed method are as followed;

Advantages

1. The DIC-ANNs detector is inexpensive.
2. The DIC lightbox is lightweight and convenient.
3. The proposed method can be used for linear and non-linear relationship between concentration and RGB value.
4. No calibration curve construction is needed, thus this method enhance rapidly of detection and low chemical consumption.

Disadvantages

1. For data processing, this software shows only the concentration input in database and could not calculate to decimal numbers in between concentration. Therefore, the sensitivity of measurement is based on the scale of calibration set used.
2. Limit of quantitation is based on color of the solution.