Implementation and Evaluation of SDS-PAGE Image Analysis on a Mobile App

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***Abstract*—SDS-PAGE is a common method in biochemistry, forensics, etc. to separate protein molecule in polyacrylamide gel. There are many tools used to analyze the gel image but most are costly and require a trained session. Smartphones have been integrated into education in aiding students to learn with visuals and friendly learning tools. This paper proposes a mobile application with capability in analyzing SDS-PAGE gel image to easily calculate the protein molecular weight anywhere and anytime. The SDS-PAGE gel image analysis on Android platform was compared between Thresholding Segmentation method on gray scale and Color Segmentation method on HSV color space. Based on the detection of gel bands, it has been found that the HSV Color Space method has fewer false negative rate than the Gray Scale method by 50%.**

***Keywords*—SDS-polyacrylamide gel electrophresis, android, mobile application, Thresholding Segmentation, Color Segmentation, gel image analysis**

I. INTRODUCTION

SDS-PAGE (Sodium Dodecyl Sulfate-Polyacrylamide Gel Electrophoresis) is a method for separating the protein molecule according to their sizes. It is commonly used for the study of the protein or peptide from the biological sample. The principle of separation is based on the molecular mesh of polymerized acrylamide that can retard the protein molecule mobilized under electrical current. The bigger size results in the slower the movement of the protein. The distance of the protein band is used to calculate for molecular weight (M_w) using protein standards of known M_w as reference [1].

There are several tools for analysis the gel image, such as Biologics Analysis Workflow[1] and Gel Documentation[2]. However, the program and the related equipment are for purchase with high price and therefore they are rarely available in general college biochemistry labs. Moreover, users cannot access such tools at anytime and anywhere.

Nowadays there are 200 million people who use smartphone (145 million for Android) [2] because they are highly capable but yet portable. In this study, we are interested in developing a mobile application for image analysis of SDS gel. This application has a high potential as a tool for each individual student to learn how to calculate M_w of a protein from their own smartphones.

In brief, we apply two approaches for image analysis. The first is Thresholding Segmentation method on gray scale which has a large variation in background intensity [3]. The two main intensities of the gel image are 1) gel band intensity and 2) background intensity. The second approach is Color Segmentation method on HSV color space. The two main colors of the gel image are 1) gel band color and 2) background color.

II. BACKGROUND

A. Related Work

COPS [3] and CODES [4] focused on Atlantic salmon protein pattern analysis. For COPS, they proposed desktop application based on gray scale image analysis. For CODES, they used gel image analysis software called AlphaImagerHP for standardized digitization and AlphaEaseFC for quantification of protein gel image. Both of them are effective for gel image analysis but inconvenient to use at anytime and anywhere. They also developed NeVPAS as a web application for storing the databanks of protein information but the users must connect to Internet.

Our work has two main contributions as following.

1. We implement and evaluate the first Android application for SDS-PAGE gel image analysis that the users can just access from their own mobile devices for work at anytime and anywhere and also available when offline.
2. We compare the performance between Thresholding Segmentation method on gray scale and Color Segmentation method on HSV color space for SDS-PAGE analysis on Android platform. This is the first work that applies Color Segmentation method on HSV color space for SDS-PAGE analysis.

[1] http://www.bio-rad.com/en-tg/sku/170-7991w-biologics-analysis-workflow
[2] http://www.bio-rad.com/en-tg/sku/170-8270-gel-doc-ez-system

B. OpenCV4Android

OpenCV4Android is an open source library used for image processing on Android platform which its library is based on Java [5].

C. Color Model

For analyzing color image there are two mostly used color models which are RGB and HSV color spaces. RGB is widely used for image color generator because human eye is strongly perceptive to red, green, blue primaries [6] but very variant to illumination condition [7]. On the other hand, HSV is widely used for developing image processing algorithms based on color descriptions that are natural to human's perception [7] and invariant to illumination condition [8].

Some works [9-11] have shown that the HSV Color Space is more appropriate for color segmentation. Thus HSV color space was chosen in Color Segmentation method for detecting gel bands.

III. METHODOLOGY

We apply segmentation method to find the gel bands. Thus, we are interested to take an initiative step to analyze a gel image on a mobile device.

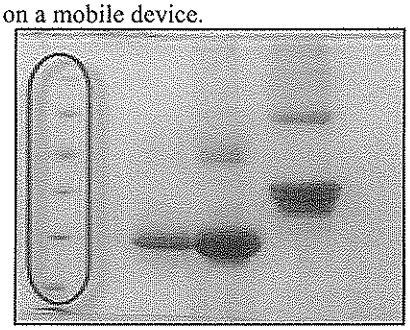


Fig. 1 An original image with standard markers in a red circle

We also use cvBlobDetection library [12] developed for detecting gel bands which are referred as "blob". It is an Android open source library that performs blob detection and extraction.

Next, we will explain Thresholding Segmentation method on gray scale and Color Segmentation method on HSV color space. To help readers to understand these methods, we will use Fig. 1-6 in our explanation. Fig 1. shows an original image with standard markers in a red circle.

A. Thresholding Segmentation Method on Gray Scale

There are five steps in Thresholding Segmentation method

Step 1: Convert the gel image to a gray scale image.

Step 2: Implement Gaussian filter to remove noise on the gel image with 5x5 mask as shown in Fig. 2.

Step 3: Increase the contrast of the gel image.

Step 4: Apply Thresholding Segmentation to separate the gel bands from the background by obtaining a threshold value from the histogram as shown in Fig. 3.

Step 5: Find the gel bands by using cvBlobDetection. Fig. 4 illustrates the result of gel band detection. The bands will be shown as color (green or blue) and then have the red rectangle around them.

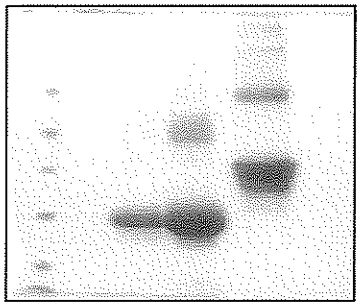


Fig. 2 Gel gray scale image

B. Color Segmentation Method on HSV Color Space

There are four steps in HSV Color Segmentation method

Step 1: Convert the gel image to HSV Color Space as shown each component in Fig. 5.

Step 2: Plot the H-S graph as shown in Fig. 6.

Step 3: Choose the appropriate H and S range of the gel band from H-S graph by using H-S graph that we will discuss in the next section.

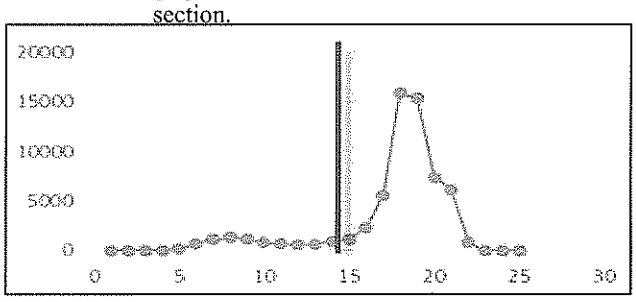


Fig. 3 Histogram of Fig. 2

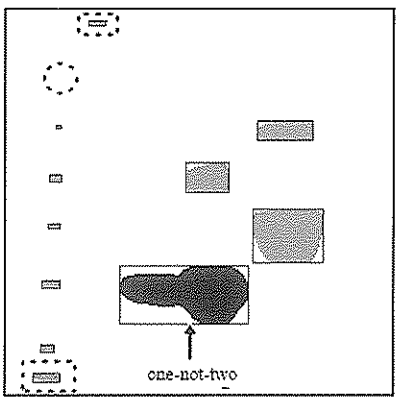
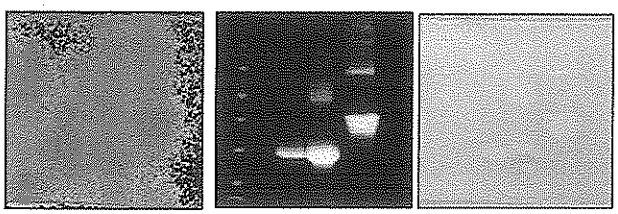


Fig. 4 The result of applying Thresholding Segmentation and band detection

Step 4: Find the gel band by using cvBlobDetection.



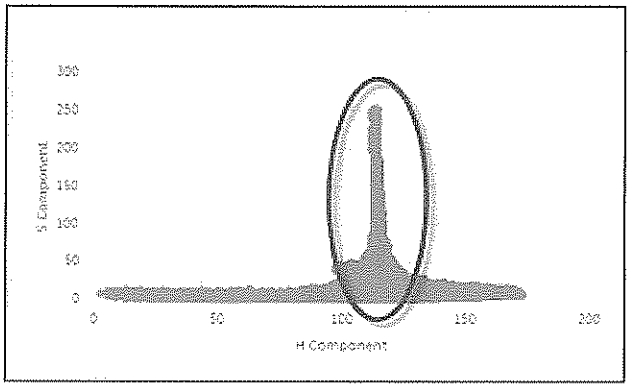
(a) (b) (c)

Fig 5 a) H component b) S component c) V component

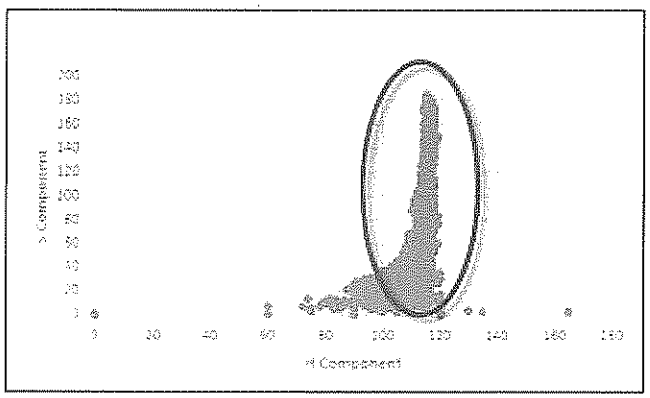
Fig. 5 shows that S component can clearly separate the gel bands from the background. The band area is white while the background area is black. We find the optimal range of H and S by plotting the graph as shown in Fig. 6.

Fig. 6a) shows that the H-S graph of Fig. 1, and Fig. 6b) - 6c) illustrates those of some other images. In these pictures, the optimal H and S ranges are in red circles.

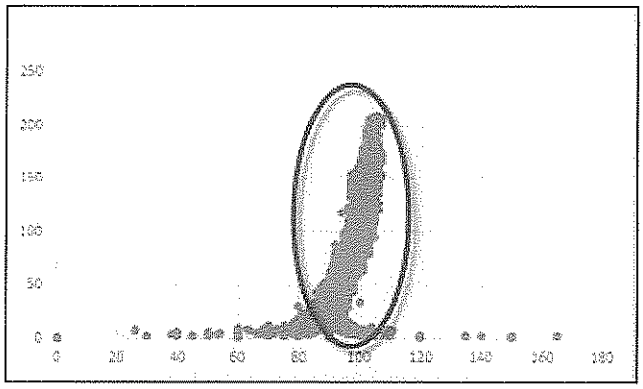
Fig. 7a) shows the image obtained from applying Color Segmentation method. The background area is black and the gel bands area is white. Fig 7b) shows the result of gel band detection with the red rectangle around each band.



(a)



(b)



(c)

Fig. 6 a) H-S graph of fig. 1 b) and c) H-S graph of other gel images

IV. Result and Discussions

In this paper, we have studied and analyzed the SDS-PAGE's gel bands image segmentation by using Thresholding Segmentation method on gray scale and Color Segmentation method on HSV color space. The gel sample image has 11 bands (6 for standard markers) as shown in Fig. 1.

In the result analysis, we define false positive for the blob as the percentage of gel bands that are not detected and false negative for the blob as the percentage of bands that are not but detected as gel bands.

The result of Thresholding Segmentation method on gray scale is illustrated in Fig. 4. It shows that there is 1 of 11 blobs that are gel bands but cannot be detected (the area with a black dashed circle) and two blobs that are not gel bands but detected (the areas surrounded with a black dashed rectangle.)

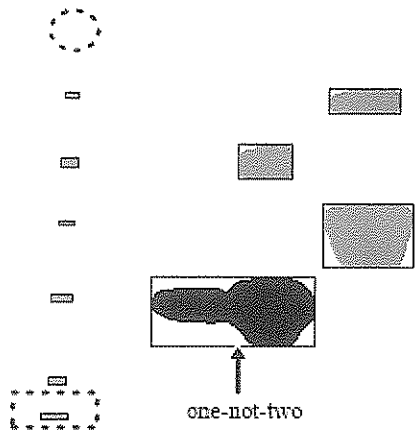


Fig 7 The result of applying Color Segmentation and band detection

The result of Color Segmentation method on HSV Color space is shown in Fig. 7. It shows that there is 1 of 11 blobs that are gel bands but cannot be detected as (the area with a

black dashed circle) and one blob that is not a gel band but detected (the area with a black dashed rectangle.)

There is one common mistake shared by both SDS PAGE image analysis methods. The mistake is shown with the label "one-not-two" which means one band is detected as two bands. These two bands are very close together so they are detected as two bands instead of one. Both methods should understand this band as one band instead of two bands.

The accuracy of band segmentation methods is shown in Table I. Thresholding Segmentation on gray scale image misinterprets some bands that have their intensity close to that of the gel bands. However, Color Segmentation on HSV Color Space can differentiate S component from the other components in the gel image. Thus for SDS image analysis, the Color Segmentation method on HSV Color Space has fewer false negative than the Thresholding Segmentation on Gray Scale.

TABLE I. ACCURACY OF BAND SEGMENTATION METHOD

Method	False Positive (%)	False Negative (%)
Thresholding Segmentation	9.09	18.18
Color Segmentation	9.09	9.09

V. CONCLUSIONS

We have developed the first Android application for SDS PAGE gel image analysis. We also have analyzed the performance comparison between Thresholding Segmentation method on gray scale and Color Segmentation method on HSV color space for SDS-PAGE analysis. The experimental analysis has revealed that Color Segmentation method yields false negative about 50% of Thresholding Segmentation method.

In this work, the result of gel band detection is shown in an area. We plan to find the real position of the gel bands by calculating the point of each gel line where the slope changes from positive to negative [3].

For the future work, we are interested in developing a hybrid approach that performs the intersection of the result images of applying both Thresholding Segmentation method on gray scale and Color Space method on HSV color space. We also plan to enhance our proposed method to reduce the false negative rate.

VI. ACKNOWLEDGEMENT

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