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Mobile Application for Biosensor Colorimetric Analysis

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Abstract— Inexpensive paper-based biosensors can be valuable screening tools to test for various illnesses, but it is often challenging to design them to produce a visual change that can easily be identified by untrained users. This research examines one method of compensating for the lack of distinct visual cues by developing and testing a mobile application that uses a machine learning algorithm (k-Nearest Neighbors) to analyze a picture of a sensor and determine whether it shows a positive or negative result. The machine learning algorithm was trained on a set of labeled sensor images and k-fold cross-validation was used to analyze its classification accuracy.

Index Terms— Classification, Colorimetric analysis, k-Nearest Neighbors (KNN), Machine learning, Mobile applications

1 Introduction

Inexpensive paper-based biosensors have the potential to be utilized as a screening tool to test for various illnesses. They would be particularly useful in situations where more traditional methods of testing are unavailable, too costly, or where there is a lack of skilled technicians to operate the equipment and analyze results. For example, a paper-based biosensor could be used to screen for malaria or dengue fever in developing nations. However, it is often difficult to design the sensors to produce an obvious visual change so that the result can be easily interpreted by the untrained eye.

It may be possible to compensate for the lack of distinct visual cues in some paper-based biosensors by software rather than by designing better "hardware" (the physical sensor itself). The most accessible and convenient software solution would be an app on a smartphone, given the capabilities of smartphones (camera, processing power, etc.) and widespread availability of smartphones all over the world. Such an app would allow the user to take a picture of the sensor, analyze the picture, and display the result to the user. The main goal is consistent, accurate result interpretation, but other goals may include simplified user interface (UI) and ease of use (UX) to make the app truly accessible to even novice users. This paper explores the feasibility of such an app, powered by machine learning, as a convenient and accessible software solution to address the difficulty in differentiating sensor results by eye.

Android was selected as the mobile platform for this app for its availability (for both users and the developer). According to the International Data Corporation, in 2015 Android had over 80% market share followed by iOS with about 14% [1]. Android devices cover the full spectrum of capabilities and price points of smartphones and are backed by many hardware manufacturers around the world while Apple only markets a few pricier, "premium" iOS products. In addition, Android Studio (IDE officially supported by Google) is available for free on Windows, Mac, and Linux whereas developing an Apple iOS app requires a Mac.

There exists some prior work on using a smartphone platform for colorimetric analysis, but at the time of writing, there are none that incorporate machine learning. Shen, et al. [2] quantified paper-based immunoassays with a smartphone by using chromaticity values to construct calibration curves of analyte concentrations and compensating for ambient lighting changes with a calibration technique using a mapping algorithm based on the measured RGB intensities of a reference chart. They report successful results with their method but assume that ambient lighting will be shining uniformly across the sensor area. Also, calibration curves are less adaptable to different sensors than a machine learning approach. Alstrøm, et al. [3] implemented K-nearest neighbor, artificial neural networks, and sparse logistic regression for colorimetric analysis but utilized a flatbed scanner to obtain images of the sensors, so they did not take into consideration how machine learning approaches will work in an environment where lighting conditions can be widely variable. Colorimetrix.com advertises a smartphone app for spectrophotometry, but states "we are very early stage," provides no demos on the website (though it is apprently "validated by experimenters in laboratories at the University of Cambridge"), and lacks details such as whether or not machine learning is used (it only mentions "an ingeniously designed mathematical algorithm"). At the time of writing, it seems no one has yet created a mobile application for colorimetric analysis that uses machine learning for classifying photos of sensor results taken by the device's camera that can work under different lighting conditions.

2 Methodology

2.1 App Development

A Samsung Galaxy S6 running Android 5.1.1 was used as the development device. The app's code uses Android support libraries and is designed to work on Android 4.1 and newer (which covers 94.8% of devices at the time of writing according to Android Developers Dashboard). Currently, the app displays an overlay of hard-coded regions over the live camera preview, allows the user to take a picture and label it POSITIVE or NEGATIVE, crops the picture to the regions indicated and averages the pixel values within each region. All camera settings are using the default profile for the phone with the exception of tap-to-focus which was customized and implemented in the app. Exposure, white balance, etc., are using the system's default algorithms and is allowed to change automatically without customization or input from user. Each sensor region information (picture, average RGB values of each region, etc.) is saved to a database on the phone (using ORMLite). The app also allows the user to view a text representation of all the data in .arff format for use with Weka machine learning library.

2.2 Biosensors

Sensors were provided by Hyun Kwon, Ph.D., at the Department of Engineering and Computer Science, Andrews University. These wax-printed, nitrocellulose sensors use antibody-functionalized gold nanoparticle probes for detection of prostate-specific antigens (PSA). When the probes bind to PSA, they remain on the sensor paper and produce a pink-red color. Schematics of this sensor are shown in Fig. 1. Due to difficulties involved in using biological samples at this stage of development of these sensors, artificial samples were applied to the sensor regions. After preparation, the sensors were laminated to prevent drying of the sensor spots and resulting color fade (another limitation of the current iteration of the sensor).

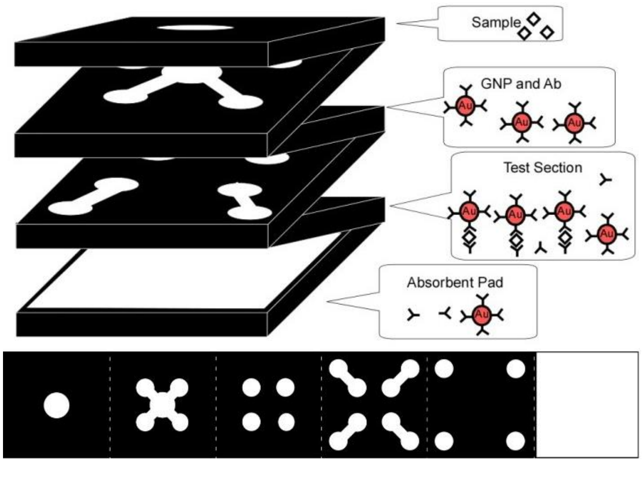

Fig. 1. Schematics of paper-based biosensor developed by Alyson Drew [4].

2.3 Data collection and Training

Five sensors, each having four distinct regions/spots, were used for traning. Each spot had applied to it 500 μg/ml, 250 μg/ml, 50 μg/ml samples or no sample (control). Data to be used as feature values in the machine learning algorithm was collected using the app. 500 and 250 μg/ml spots were labeled POSITIVE and 50 μg/ml and control spots were labeled NEGATIVE. Two pictures of each spot were taken under five different lighting conditions – Incandescent (indoors), Fluorescent (indoors), Overcast (outdoors), Sunny (outdoors), and Shade (outdoors) – for the sum of 40 data points for each lighting condition and a total of 200 over all lighting conditions.

KNN was chosen as the machine learning approach to evaluate because while it is a simple algorithm, previous research has found it to be as effective in classifying results as more complicated methods such as sparse multinomial logistic regression and artificial neural networks [2]. The average red, green, and blue (RGB) values of the pixels within the region were used as the feature values.

2.4 Analysis

The data collected was downloaded from the phone to a computer and analyzed off-line using a custom program implementing the Weka machine learning library. 5-fold cross-validation was run for each lighting condition. For each of the five iterations, one sensor served as the sample while the other four were used for training. The 5-fold cross-validation was repeated for K=3, 5, 7, and 9. 5-fold and 10-fold cross-validation was run on the collective dataset over all lighting conditions (Combined) using Weka Explorer. Again, this was repeated for multiple K values.

3 Results

The results of k-fold cross-validation are summarized in Table 1. The highest classification accuracy for any lighting condition was 87.5% (Sunny and Shade, K=3) and the lowest was 70% (Incandescent, K=3). In general, increasing K did not improve classification accuracy with the exception of Incandescent which showed an improvement at K=5 and also at K=7 (but not at K=9).

	K=3	K=5	K=7	K=9
Incandescent (indoors)	28/40 (70%)	30/40 (75%)	**32/40 (80%)**	30/40 (75%)
Fluorescent (indoors)	**34/40 (85%)**	34/40 (85%)	32/40 (80%)	29/40 (72.5%)
Overcast (outdoors)	**31/40 (77.5%)**	30/40 (75%)	30/40 (75%)	30/40 (75%)
Sunny (outdoors)	**35/40 (87.5%)**	33/40 (82.5%)	30/40 (75%)	32/40 (80%)
Shade (outdoors)	**35/40 (87.5%)**	35/40 (87.5%)	35/40 (87.5%)	33/40 (82.5%)
Combined (5-fold)	**170/30 (85%)**	166/200 (83%)	164/200 (82%)	166/200 (83%)
Combined (10-fold)	**173/200 (86.5%)**	168/200 (84%)	163/200 (81.5%)	165/200 (82.5%)

Table 1. Tabulation of correctly classified samples under each lighting condition with varying K value. The highest classification accuracy within each lighting condition is bolded.

4 Discussion

Due to the small size of the training data set (32 data points for each lighting condition), increasing K did not usually improve classification accuracy – as K gets larger, the model becomes more susceptible to noise in the training data. Also of note is that for Incandescent samples, there was a bigger discrepancy between subsequent picture of the same sensor spot compared to samples for other lighting conditions due to the influence of a secondary (dimmer) light source (non-incandescent) on the first of the two pictures for each spot. This may explain why Incandescent had the lowest classification accuracies out of all the lighting conditions across many K values.

Future work on this app includes collecting additional training data from more sensors. The sensors used in this research only had regions where sample concentrations of 500, 250, 50, and 0 µg/ml were applied, and the POSITIVE and NEGATIVE spots were easily distinguishable by eye. More gradations in the concentrations were not possible at this time. It would be valuable to explore how well KNN works on data with subtler differences between the POSITIVE and NEGATIVE classes. It would also be prudent to test other types of biosensors, particularly ones that use real biological samples rather than artificial ones.

Further, other machine learning approaches could be implemented and evaluated to compare their classification accuracy. Other methods may perform significantly better than KNN in this context and some would allow for quantitative analysis (give a predicted concentration value) besides just binary classification. Also, a computer vision library such as OpenCV could be used for automatic detection of sensor regions based on their color, shape, and size. This would greatly simplify the use of the app for consumers and the process of collecting training data.

The current version of the app was designed mainly with the researcher in mind and has only been tested on the development phone. Prior to commercialization and release to the public, the app should be tested on more devices including those running older versions of Android in order to ensure compatibility and that the app is free of bugs. Also, adding features such as automatic updates of training data, ability to report incidence statistics to a central database, etc. could transform this app from a reader to a powerful instrument for epidemiology.

Machine learning approaches work well with noisy data and thus has great potential for application in mobile apps for colorimetric analysis of paper-based biosensors where varations in lighting can contribute significant noise to input data. Further research in this area is important if these sensors – very promising in themselves due to low cost of production and portability – are to be truly useful and effective screening tools in the real world.

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Eui Bin You is a senior undergraduate student at Andrews University majoring in Computer Science. He is interested in the application of computer science, particularly machine learning and aritificial intelligence, in healthcare to improve the field of medicine.

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