Applying an Image Technology to Estimates Values of Nitrite in Processed Meat Products

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***Abstract*—Potassium nitrite or saltpeter is used as a food additive and preservative. It confers a fresh and appetizing appearance to food when used in moderation. However, when used in excess, it may lead to cancer. In the present study, an image-processing mobile application was developed for quality control and ensure the hygiene of food products. The developed application is a user-friendly innovation that would raise the quality standards of processed foods, allowing for a competitive edge in the market. The main objectives of the present study were to identify the representatives of each class of suitable color tones and then develop a model-based application for estimating the content of nitrite in processed meat products. The study was conducted in six steps: (1) image layer separation of RGB to three layers comprising the R-G-B layers; (2) identification of the representatives of each class of suitable color tones using the k-means clustering technique; (3) deciphering the linear equations representing the linear relationship between the color tone and the content of nitrite; (4) designing of a mobile application for estimating the amount of nitrite based on an image; (5) development of the model-based mobile application for estimating the nitrite content; (6) evaluation of the developed mobile application using the testing dataset. The results revealed that the mean and median of the green color's layer were appropriate representatives of the image dataset and could also be associated with the concentration of the nitrite standard solution. In addition, the efficiency of estimating the concentration of nitrite in meat products using the paper analytical apparatus was 88.25%.**

***Keywords*—image processing, mobile application, linear equation, k-means clustering, microfluidic paper-based**

I. INTRODUCTION

A suitable innovation for food quality control would raise the quality standards and competitiveness of food products. The innovative technology used for quality control and ensuring good hygiene of food products exerts a direct positive impact on the value of the food products. In Nakhon Ratchasima province (Thailand), which is commonly referred to as Korat, processed meat products are consumed as traditional foods of the local culture of Korat and are usually referred to as "mam". These products include sausages prepared using animal liver, beef, or pork. The community that prepares these processed foods are scattered across different districts of the Nakhon Ratchasima province, due to which "mam" is considered a community product. The majority of these community entrepreneurs preparing "mam" sell their products on roadside stands, for which the products are hung on a sun rack to accelerate the fermentation process. This hanging fermentation process exposes the products to dust, germs, and various environmental pollutants, leading to low standards and a lack of hygiene in these products. Therefore, it is important that the large community of entrepreneurs involved in the preparation of "mam" are encouraged to establish suitable quality and hygiene standards for their food products to ensure a competitive edge in the market for these products. This would also contribute to creating more jobs and strengthen the local economy in the community.

Plungklang *et al*. [1] conducted a general survey of the entrepreneur community in the Nakhon Ratchasima province. The project included 26 community entrepreneurs in four districts of the province, namely Chakkarat, Muang, Nonthai, and Kham Talay So, and the objective was to promote the quality control of processed meat food products prepared by these community entrepreneurs. In the same context, the present study aimed to develop a mobile application to determine the nitrite content in processed meat products to resolve hygiene-related concerns and raise the quality standard of these community products. The proposed application is designed to serve as a low-cost, easy-to-use tool aligned with the community's potential. The study also revealed

Manuscript received March 3, 2023; revised May 23, 2023; accepted July 11, 2023; published October 20, 2023.

doi: 10.12720/jait.14.5.1088-1095

that 9 of 26 entrepreneurs admitted the addition of nitrite to their products (see Fig. 1).

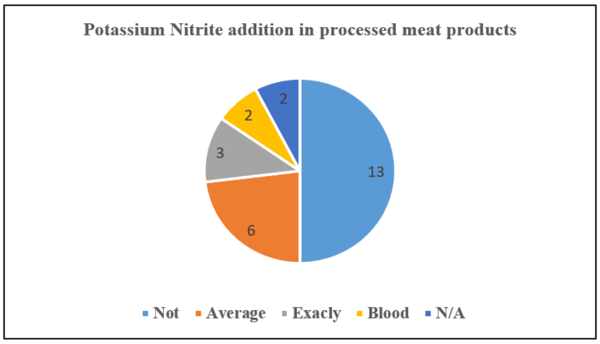

Figure 1. The nitrite addition in processed meat products [1].

Fig. 1 illustrates the widespread use of nitrite, which is reported to be harmful.

A mobile application is software that works on portable communication devices, such as mobile phones, which run on different Operating Systems (OS) [2, 3]. An application software is a program that is designed to perform a specific function to directly achieve the completion of a specific task [3], such as the various mobile applications for the Android platform [4], the mobile application to enhance the English language skills of hotel staff [5], and the application for managing the transportation of the elderly to the hospital in a sub-district municipality [6], among others. In image analysis, machine learning concepts are frequently applied for image processing to increase the efficiency of image analysis, such as analyzing breast cancer images [7]. In addition, these concepts have been applied to determining the level of water in plain sight using an application for detecting water bodies [8] and resolving super-resolution image analysis problems based on the deep learning approach [9].

In this context, the machine learning concepts may also be applied to develop an innovative smartphone application to monitor the nitrite levels in a food product, which would allow community entrepreneurs to control the nitrite levels in their food products. The publication of product quality control reports and public relations activities conducted for the general public would then increase the credibility of community products with a sense of confidence.

II. Literature Review

A. Nitrite in Processed Meat Products

Nitrite is a source of nitrosamine compounds, which are carcinogenic to several animals. High doses of nitrite in animals result in changes in red blood cell metabolites (such as methemoglobin), rendering red blood cells to be unable to carry oxygen for use in the body. The lack of oxygen in the cells of the body is consequently fatal [10, 11]. Therefore, announcement no. 281 of the Ministry of Public Health (2004) established the maximum allowable amount of potassium nitrite (KNO_2) in food products at the threshold of 125 milligrams per kilogram of food [12].

B. Microfluidic Paper-Based Analytical Devices (µPADs)

The microfluidic paper-based analytical devices (µPADs) were developed to evaluate the content of nitrite (NO_2) in processed meat products based on the application of the Griess reaction, which is presented in Fig. 2 [1]. The devices were developed using the following steps:

- Paper devices were fabricated using the wax screen-printing technique [13] and comprised hydrophilic and hydrophobic regions. The devices were designed to have a reaction zone for nitrite and Griess reagent. This reaction zone was a hydrophilic region of 7 circles, each with a diameter of 1.7 centimeters. The upper 4 circles were the regions that facilitated the reaction of 1% Griess reagent solution with the respective nitrite standard of concentration 0, 25, 62.5, and 125 milligrams per kilogram of feed (mg/kg). The lower 3 circles allowed the reaction of 1% Griess reagent with the 3 replicates of the sample extraction solution, namely, S-1, S-2, and S-3, respectively (Fig. 2).

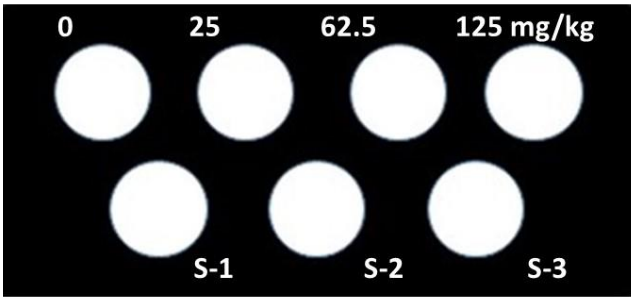

Figure 2. The developed paper analytical device, white areas represent the hydrophilic regions and black represents the hydrophobic regions. There are 7 hydrophilic circles with the same diameter of 1.7 cm.

- Nitrite level evaluation was performed using the paper analytical equipment, for which standard nitrite solutions of concentrations 0, 25, 62.5, and 125 mg/kg of food were prepared, which were equivalent to the concentrations of 0, 10, 25, and 50 mg of nitrite per liter, respectively. Next, 20 µL of 1% Griess reagent was added to the center of each of the 7 circles followed by allowing the reaction for 10 min until the reaction solutions were completely dry. Afterward, 20 µL from each of the 4 concentrations of the standard solution (0, 10, 25, and 50 mg nitrite per liter) was added to the center of the 4 upper circles, followed by the addition of 20 µL of the sample extract to the center of the 3 lower circles (S-1, S-2, and S-3). After allowing the reaction to occur for 10 min, during which all 7 reaction zones appeared pink, the intensity of the pink color was determined using the Microsoft Photoshop program. The color intensity was proportional to the amount of nitrite present in the starting solution.
- Standardized graphs were generated, and the nitrite content determined using the paper analytical equipment was calculated based on the

intensity values of the pink color that appeared at the reaction zones of the 7 circles. Next, the huge difference between the nitrite concentration and ΔH (pink intensity due to 4 among 4 circles using the above standard solution) was calculated, and the associated graphs for the relationship were plotted. The average intensity of the pink color of the sample solution (obtained based on the intensities of the pink color of the sample solutions in triplicate from the 3 lower circles) was used for calculating the concentration of nitrite in the meat processed food samples through comparison with the standard graph.

- The packaging design for the paper kit for nitrite analysis was formulated, which included creating a manual and a video demonstrating the complete set of equipment ready for field visits, dissemination, and campaigning for quality control activities. The packaging design also provided information on developing an application for a smartphone, which could be used in computer operations to further develop the application.

C. Mobile Application

Mobile applications are divided into two related parts: mobile or portable communication devices are based on telephones and work like computers. A portable device has the outstanding feature of being small, lightweight and uses relatively low power. Nowadays, portable devices can perform many functions, such as exchanging information with computers and, most importantly, can increase their work functions. Application or software assists the work of the user. The application must have a User Interface (UI) to act as an intermediary for various applications. Therefore, mobile application means an application that helps users work on portable communication devices such as mobile phones. These applications will run on different Operating Systems (OS). Examples of mobile operating systems include Microsoft's Windows mobile, Apple's iOS, and Google's Android OS [2, 3].

Mobile applications comprise software that runs on a mobile device and performs certain tasks for the user. The rapid development of information and communication technology has led to the emergence of easy-to-operate, user-friendly, affordable, and downloadable applications, which may be run on most mobile phones, leading to the widespread use of these applications for various tasks, such as calling, messaging, browsing, chatting, communication, social networking, audio, video, gaming, etc. These mobile applications are designed to allow for a further convenient daily life through connection to the internet, improved interaction with the world, better information reception, and enhanced social communication via platforms such as Facebook and Twitter. The challenges of a mobile application are its platform capability and limitations, such as small screen size, lack of windows, navigation, and speed. Despite these limitations, the uses and popularity of mobile applications are increasing with time, with a majority of people preferring mobile devices and mobile applications over desktops for simple tasks. Consequently, all major mobile manufacturing companies and mobile application developers are increasing their capacity, quality, and functionality, rendering modern mobile applications further capable and user-friendly, resulting in an all-time high global impact [14].

D. Image Technology

Several tools and methods are available for extracting key image characteristics. Among these methods, certain interesting ones include the use of color tone and intensity as features for classification. The intensity and the color tone that appear in the image represent the essential features of the image and may reflect the similarities/differences among images [15].

Several color models are in use currently, and their selection depends on the implementation. However, generally, all standards follow the same concept of representing the colored dots within the 3D grid. The colors that are usually observed appear when the light strikes an object and then reflects toward human eyes, causing humans to visualize the different colors of the object. The working principle of the computer is similar, just that the image is stored in the RGB or primary color format, with values in the range of (0 to 255) or 8 bits of each color. When the properties of the primary colors are combined, the desired color is obtained. Therefore, if one wishes to sort objects using color as the sorting criterion, one could use image processing to facilitate sorting along with the use of image data [16, 17]. The color tone and the color intensity of the image could be used together to classify digital images. For instance, the quality of mangoes was determined using a range of color values for classification [18]. In addition, the color-tone similarity of digital images [19] was used based on the color space transformation of RGB to a new format, which allowed for determining the similarity, tone, etc.

Rady *et al*. [20] applied RGB color imaging coupled with machine learning algorithms to detect plant and animal adulterants in the range of 1% to 50% in minced meat. The authors reported that gray-level and co-occurrence features were further effective compared to the other color channels when constructing the classification and regression models. The stated study provided a non-invasive and low-cost system for detecting adulteration in minced meats.

Lopes *et al.* [21] studied the automatic evaluation of raw ham quality based on image analysis, which had certain challenges due to the use of traditional Computer Vision Systems (CVS). In order to improve the CVS classification performance, the authors proposed Dual Stage Image Analysis (DSIA) as an additional step in a CVS. The classification accuracy was improved by over 20%.

Hanggara *et al.* [22] studied the fat content in beef, as fat is one of the determining factors for beef quality. While beef fat is visible on the surface of the meat cut with thin lines or in spots on the cut surface of beef, determining the beef quality level is quite a complex process. Therefore, an Android-based application based on the Gray Level Co-Occurrence Matrix (GLCM) Feature Extraction and the K-Nearest Neighbors (KNN) algorithm was developed by the authors to determine beef quality. The experimental results revealed a classification of the grade or quality of meat with the best accuracy of 91.7%.

Similarly, the present study aimed to identify a hybrid technique that would allow for creating an effective application for the benefit of the community. The search for such a technique included several important tasks. The first task was to study image classification in the RGB Model for the extraction of the key features from the image by separating the color layers as R-G-B, followed by using each color layer to identify an appropriate tone representative of the target class using the k-means clustering technique. This clustering technique measures the purity and accuracy, thereby allowing the assessment of the clustering against the true class of data. In addition, a silhouette was used, which is a clustering measure that evaluates the nature of the color tone of the image (without knowing the true class). Next, an effective classifiable agent was obtained, based on which a linear relationship between the color agent and the nitrite content was deciphered. The resulting model was then used for predicting the nitrite content on an application.

III. Materials and Methods

A. Dataset Description

The training and testing datasets were obtained from the report published by Pluangklang *et al.* [1], in which the authors had reported the results of the survey of 26 community entrepreneurs in four districts of Nakhon Ratchasima province, namely, Chakkarat, Muang, Nonthai, and Kham Talay So, with the aim of promoting the quality control of processed meat products in this region. The images in the testing set had a resolution of 135×122 pixels, with a total of 468 images classified into the following four classes: class 0 mg/kg, class 25 mg/kg, class 62.5 mg/kg, and class 125 mg/kg. Each class presented a color scheme in the microfluidic paper-based analysis conducted to allow a reaction of 1% Griess reagent solution with the respective nitrite standards at the concentrations of 0, 25, 62.5, and 125 milligrams per kilogram of food (mg/kg). The examples are presented in Fig. 3.

Figure 3. Examples of images used for testing set data consist of image (a) class 0, (b) class 25, (c) class 62.5, (d) class 125.

B. The Proposed Work

The work proposed in the present study included the following steps:

- Image layer separation of RGB into three layers comprising the R-G-B layers followed by converting each layer into color vectors.
- Image analysis to identify the representatives of each class of suitable color tones among the images using the k-means clustering technique.
- Generation of a linear equation representing the linear relationship between the color tone and the amount of nitrite.
- Analysis and designing of a mobile application for estimating the amount of nitrite from an image.
- Developing the mobile application for estimating the nitrite content based on the linear equation.
- Evaluating the developed mobile application using the testing dataset.

C. Key Feature Extraction

The proposed representative indices were identified from the R-G-B layers using the representatives of each color layer, based on which key feature extraction was performed for each layer. The dataset used in this experiment was separated into three layers, with each layer containing a vector of 16,470 dimensions, i.e., a high-dimensional vector. Therefore, the experiments of the present study were also conducted under dimension reduction. The appropriate representative from each color layer was used. The mean and median values of each layer of each image were generated.

The color-tone distribution of the dataset was classified according to the target class based on the mean of each class (R, G, and B), as illustrated in Fig. 4.

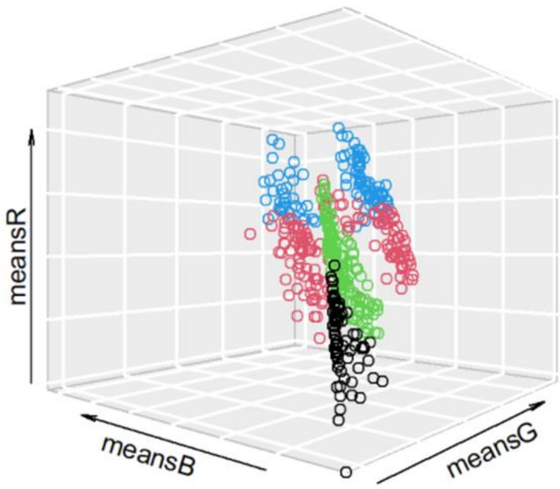

Figure 4. The distributed clusters based on the color tone for each layer in RGB mode.

D. k-Means Clustering

The k-means clustering technique relies on the center of the cluster, with the cluster centroid often represented by the mean of the cluster. In the clustering process, the similarity of the group is measured by iterating the measured distance between each object and the center of each cluster [23] using a Euclidean distance measure.

The k-means algorithm is, accordingly, described as follows [24, 25]:

- Choose initial centroids $\{m_1, \ldots, m_k\}$ of the clusters $\{C_1, \ldots, C_k\}$.
- Calculate new cluster membership. A feature vector x_j was assigned to the cluster C_i if and only if.

$$i = \underset{k=1,\dots k}{arg\ min} \left\| x_j - m_k \right\|^2 \quad (1)$$

- Recalculate centroids for the cluster according.

$$m_i = \frac{1}{|C_i|} \sum_{x_j \in C_i} x_j \quad (2)$$

- If none of the cluster centroids have changed, finish the algorithm. Otherwise go to Step (2nd).

IV. Experimental and Result

A. Clustering Experiments

These experiments were conducted to analyze the image and identify the appropriate key features that could be used to represent the image and also to indicate the class target. In the present study, the stratified R-G-B image dataset was used and analyzed using unsupervised learning techniques such as k-means clustering. The k-means clustering technique involves determining the nature of the similarity of data and grouping similar data into the same groups [20]. In the present study, the k-means algorithm was applied to allow learning the nature of the dataset, followed by grouping and evaluating the results of the clustering. In total, 14 cases were designed, and the details of each case are presented in Table I.

TABLE I. Description of Case for k-Means Clustering

Case	Name	Description
1	All-Layers	Using all layers of image
2	Mean R-G-B	Using mean of each layer R-G-B
3	Mean G-B	Using mean of each layer G and B
4	Mean R-B	Using mean of each layer R and B
5	Mean R-G	Using mean of each layer R and G
6	Median R-G-B	Using median of each layer R-G-B
7	Median G-B	Using median of each layer G and B
8	Median R-G	Using median of each layer R and G
9	Median R-B	Using median of each layer R and B
10	Median R-G-MeanR-G	Using median of each layer G and B, and mean of each layer R and G
11	Median R-G-Mean G	Using median of each layer R and G, and mean of layer G
12	Median G-Mean G	Using median of layer G, and mean of layer G
13	Median G	Using median of layer G
14	Mean G	Using mean of layer G

Then compare the results of the clustering between accuracy, purity, and silhouette, which the grouping results show as shown in Table II.

Table II presents the representatives with good clustering results. The high accuracy and purity values indicated that the clustering corresponded to the true class of the image. The high silhouette values indicated that the clustering corresponded to the natural color tone of the image. If all 3 values are consistently high, then the case representative is considered suitable for use as a predictor. In Table II, the top 2 results are highlighted, with case 13 as a good representation of all three values.

TABLE II. The Comparing of k-means Clustering Results

Case	Representatives	Purity	Accuracy	Silhouette
1	All-Layers	66.88	67.05	0.59
2	MeanR-G-B	66.03	66.23	0.58
3	MeanG-B	68.38	68.47	0.61
4	MeanR-B	55.77	57.02	0.64
5	MeanR-G	72.44	72.54	0.68
6	MedianR-G-B	70.73	71.00	0.58
7	MedianG-B	**87.18**	**87.01**	0.68
8	MedianR-G	74.77	74.89	0.68
9	MedianR-B	65.38	66.69	0.59
10	MedianR-G-MeanR-G	74.15	74.30	0.68
11	MedianR-G-MeanG	75.43	75.58	0.70
12	MedianG-MeanG	77.35	77.41	**0.74**
13	MedianG	**79.27**	**79.33**	**0.75**
14	MeanG	***75.64***	***75.60***	**0.75**

B. Best Representative of the Dataset

The results obtained in the determination of a suitable color tone were representative of the image dataset of paper μPADs by applying the clustering technique to the test image dataset with the standard substance with a known nitrite value (called target set). It was revealed that the best representation of the tones from the image was the median of the green layer, which had the highest purity and accuracy values of 79.2735 and 79.32726, respectively (Fig. 5). The suitable silhouette value was determined to analyze the image's nature while ignoring the original data. It was revealed that the most appropriate representation of the color tones from the images was the mean of the green layer, which presented the highest value of 0.75 (75%), as depicted in Fig. 6.

```
##
##      0  25 62.5 125
##   1  0   0   2  78
##   2 24  98  13   0
##   3  0   1 102  39
##   4 93  18   0   0
```

```
Prt <- ClusterPurity(kmean.result$cluster, ClassSaRGB$class)*100;
print(Prt)
```

```
## [1] 79.2735
```

```
Acc <- cluster.evaluation(ClassSaRGB$class, kmean.result$cluster)*100;
print(Acc)
```

```
## [1] 79.32726
```

Figure 5. Evaluate the k-means clustering of targets set with purity and accuracy value.

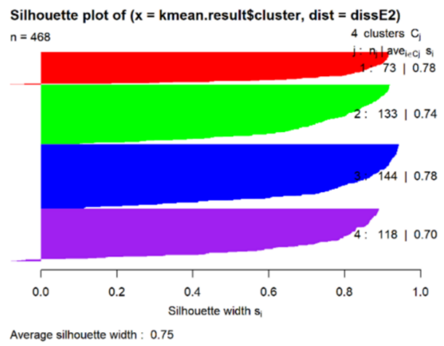

Figure 6. Evaluate the k-means clustering of targets set with silhouette value.

C. Linear Equation

In the image analysis conducted to estimate the nitrite content in the present study, a linear relationship between the nitrite content and the representative value of the target image dataset (color tone representative) was used as a model in the application. The correlation between the nitrite content and the color tone representative reflected the following two relations:

- The first relationship was a correlation between the nitrite content and the median of the green layer of each image, which could be expressed as a linear equation in terms of Eq. (3).

$$x_1 = \frac{medianG - 0.7963}{-0.0024} \tag{3}$$

The analysis of the relationship between the tone representative (median of the green layer) and the amount of nitrite revealed a linear relationship equation, presented in Fig. 7, with an R^2 value of 0.9723.

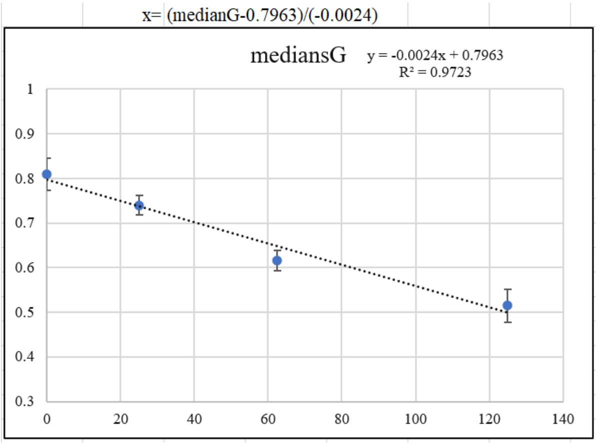

Figure 7. Linear relationship between median G and nitrite intensity.

- The second relationship was the correlation between the nitrite content and the mean of the green layer of each image, which could be expressed as a linear equation in terms of Eq. (4).

$$x_2 = \frac{meanG - 0.7954}{-0.0022} \tag{4}$$

Similarly, the analysis of the relationship between the mean of the green layer and the amount of nitrite revealed the linear relationship equation presented in Fig. 8, which had an R^2 value of 0.9685 (Fig. 7).

- In order to obtain the most accurate prediction, used the average value of nitrite content estimated from the above two equations was used in the present study (Eq. (5)).

$$\bar{x} = \frac{x_1 + x_2}{2} \tag{5}$$

when,

x_1 is the estimated amount of nitrite from Eq. (3).

x_2 is the estimated amount of nitrite from Eq. (4).

$\bar{x}$ is the average amount of nitrite from Eq. (5).

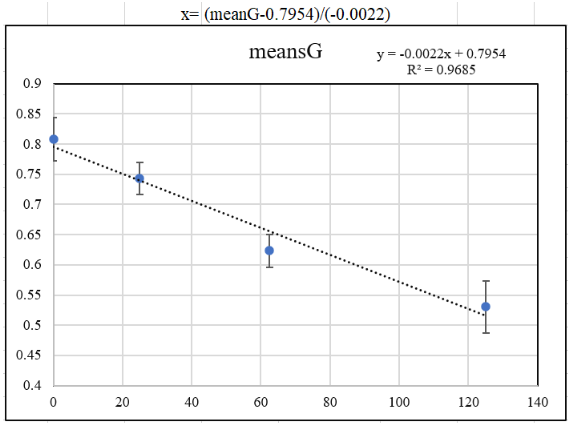

Figure 8. Linear relationship between mean G and nitrite intensity.

D. Analysis and Design of Mobile Application

The results of the analysis and design of a mobile application to estimate the content of nitrite in processed meat products revealed a system working process presented in Fig. 9, which comprised the following 7 steps:

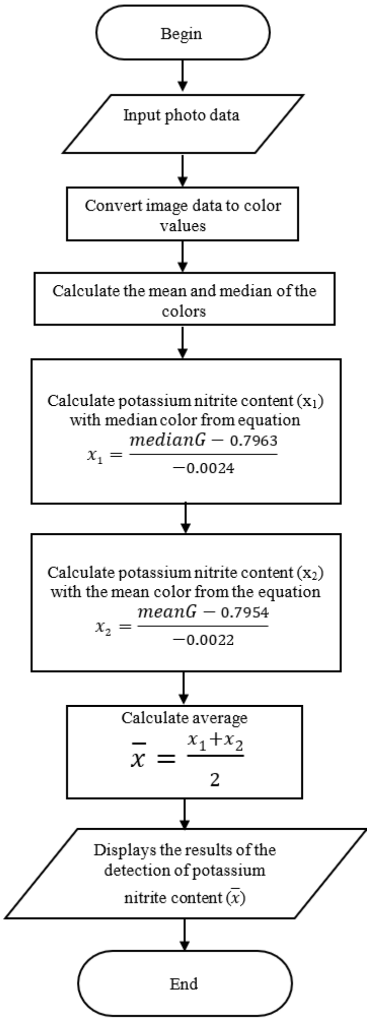

Figure 9. An operating of application are 7 steps.

- Input the image data;
- Convert the image data to color values;
- Calculate the mean and median of the colors;
- Calculate the nitrite content (X_1) with the median color;
- Calculate the nitrite content (X_2) with the mean color;
- Calculate the average nitrite content;
- Display the results of the determination of nitrite content.

E. Development of the Mobile Application

The development of the mobile application to estimate the content of nitrite in processed meat products was performed next in the present study. The application could obtain µPADs images and process the images to estimate the content of nitrite in the meat products, as depicted in Figs. 10 and 11.

Figure 10. Main screen.

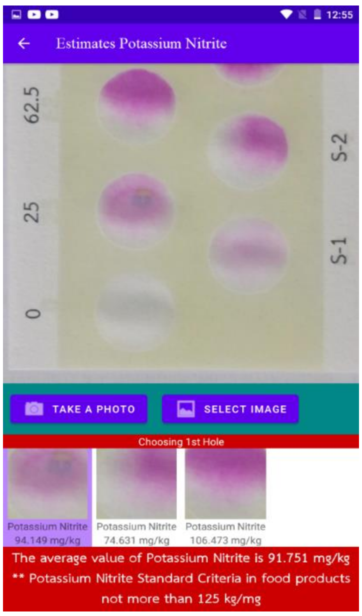

Figure 11. Example of nitrite estimate result.

F. Evaluation of the Developed Application

The final step in the present study was to evaluate the efficiency of the developed mobile applications in the estimation of nitrite content in processed meat products using the test paper µPADs. The dataset on which the application was tested included images with a size of 135×122 pixels. The test dataset included a total of 468 images that were classified into four groups containing 117 images each and respectively representing class 0, 25, 62.5, and 125. According to the results, 413 images among the evaluated 468 images could be classified with an accuracy percentage of 88.25%.

V. Conclusion and Prospects

A. Conclusion and Discussion

The results of the present study may be summarized as follows:

- The median value of the green layer and the mean value of the green layer were suitable representations of the image data.
- The analysis of the relationship between the color tone of the image and the amount of nitrite revealed two linear relationships in terms of Eqs. (3)–(4), and the average nitrite content was estimated using both the above equations.
- The developed mobile application could obtain the images of test paper µPADs and then process these images to estimate the content of nitrite in processed meat products.
- A nitrite estimation efficiency analysis of the developed mobile application revealed that among the evaluated 468 images, 413 images were estimated correctly, accounting for an accuracy percentage of 88.25%.

According to the above result summary, it may be stated that the present study relies on the analysis of image data in the RGB model, similar to the studies of Rady *et al.* and Hanggara *et al.*, with the exception that the present work did not require transforming the data to grayscale. A review of the studies on the optimization of data classification revealed that almost all the reviewed studies attempted to present the representation method. The present study used the k-means clustering technique and obtained the suitable representatives of the color tone, which were the mean of the green layer and the median of the green layer. This was different from the other works, such as the work of Lopes *et al.*, in which the split-region of interest technique was used, and the CVS classification with Dual Stage Image Analysis (DSIA) was improved, or the study of Hanggara, which relied on GLCM extraction.

The final prediction results of the present study achieved a prediction accuracy of 88.25%, which was lower than that reported by Haggara *et al.* and higher than that reported by Lopez *et al.* Haggara *et al.* reported an accuracy of 88.10%, with R^2 values in the range of 96.85%–97.23%, which is close to the work of Rady *et al.*, who reported accuracy values ranging from 88.00% to 98.00%.

In addition, the present study developed an application that is convenient and rapid to operate and did not consume the resources of a mobile device as in Rady *et al*.'s work.

B. Prospects

According to the results of the present study, the representation of the image dataset obtained through the clustering technique may not have much accuracy in classification. Therefore, to improve the accuracy of estimation, researchers should explore other techniques for training artificial intelligence machines to learn and classify images further accurately.

Conflict of Interest

The authors declare no conflict of interest.

Author Contributions

T. T., J. R., and M. T. collected, stored, and prepared data developed by the system for the estimated value of nitrite in processed meat products; analyzed and designed a system for estimating the value of nitrite in processed meat product; analyzed, summarized, and discussed the research results, and written research articles for publication in journals. T. P., V. C., C. L., M. S., P. P., and C. P. studied and researched the measurement of nitrite in meat products, developed a set of Microfluidic paper-based analytical devices (μPADs), and reviewed research article content, all authors had approved the final version.

Funding

This research was supported by the strategy budget of Nakhon Ratchasima Rajabhat University (NRRU) and has been funded by NRRU.

Acknowledgment

The authors appreciate the financial assistance provided by Nakhon Ratchasima Rajabhat University. Thank you to the Information Technology Program, Computer science program, and Chemistry program for research support.

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