Rapid Testing of Aflatoxin by Using Image Processing and Artificial Neural Network

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Abstract. Aflatoxin can be recognized clearly by using UV-light. This information is very important to develop the device for detecting the aflatoxin inside the corn by using image processing. Current research related to identification of aflatoxin has been conducted manually by the experts. This method have some weakness including subjectivity factors, inconsistent result, and time required used. Based on the problems above, it needed to create the rapid testing device for identification of aflatoxin with consistence result, accurate and easy to operate. The research aimed to develop the device for rapid testing of aflatoxin. The method used image processing and artificial neural network. The raw material used was the corn. The image of aflatoxin taken by using digital camera (Gopro 4) and processed by image processing program. ANN model was developed with 10 input parameters, 20 hidden layers and 4 targets. The fourth of targets above were the size of aflatoxin such as 1 - 2 ppb, 2 – 3 ppb, 3 – 4 ppb dan 4 - 5 ppb. The result showed that the characteristics of image were very specific among the input parameters and the most influential to recognize the object was the longest diameter of aflatoxin. The result of training showed that the size of aflatoxin can be recognized by the system was 100%, while the validation by using the different sample was 99%. Based on this research can be conclude that image processing can be used as the rapid testing of aflatoxin.

1. Introduction

Aflatoxin is a naturally occurring toxin produced by the fungus Aspergillus flavus. The fungus can be recognized by a gray-green or yellow-green mold growing on corn kernels in the field or in storage plant stress due to drought, heat or insect damage during fungus growth usually increases aflatoxin levels. Since aflatoxins are strongly carcinogenic in humans and domestic animals, it is important to be able to reliably analyze foodstuffs for their presence [1][2][3].

For the above reason, several methods for detecting aflatoxins in agricultural commodities have been reported: thin layer chromatography (TLC), high-performance liquid chromatography (HPLC) with fluorescence detection, HPLC with mass spectrometry, and the use of rapid and simple enzyme linked immune sorbent assay (ELISA)-based aflatoxin screening kits, which are commercially available for all major types of aflatoxin [4][5][3][2][6]. However, these methods generally require considerable time for sample preparation, and use expensive disposable columns for clean-up, such as the multifunctional column (MFC) and the immune affinity column (IAC).

Aflatoxin contaminated corn may show a bright greenish yellow fluorescence (BGYF) under ultraviolet (UV) light at a wavelength of 365 nm. Traditionally, BGYF positive corn are manually separated by workers. However, manual selection depends on the expertise level of the workers and it may cause skin-related health problems due to UV radiation. The technology of digital image processing
and artificial neural network (ANN) can be an alternative option to overcome the problems of identification of corn quality. The operational of this technology is very simple and fast in processing system. The accuracy level can be trusted, and the system is relatively inexpensive with simple equipment. The most complex system is the software, which requires an understanding of digital image processing and ANN which then formed into the programming system. This method is more sensitive as it is equipped with electro-optics sensors. This sensors could definitely be more precise when compared with human visual provide good information when combined with a decision support system with high accuracy that highly influenced by the psychological condition [7]. Image processing can provide good information when combined with decision support system with high accuracy. Kusumadewi [8] suggests that the use of ANN will enable optimal results, because it has advantages in solving the non-linear problems.

The technology of digital image processing has been applied widely in the agricultural problems, such as determination of fruit quality by size, shape and color. Evaluations of quality of some agricultural commodities using image processing and ANN have been reported elsewhere, such as sorting system for star fruit [9], physical quality of coffee beans [10], edamame [11] and cut flowers [12]. Such studies are very useful in identifying quality of commodity and it is the basis for research and development of non-destructive evaluation. The research aimed to develop the device for rapid testing of aflatoxin. The methods used image processing and artificial neural network. It is expected from this study that it can overcome the problems of aflatoxin contamination on the corn commodity.

2. Methods
The study was conducted at the Indonesian Center for Agricultural Postharvest Research and Development, Bogor, Indonesia. Research of rapid testing for aflatoxin in corn by using image processing and artificial neural network. The step of research consisted of generating data, training, validation, and creating the application program. The method used was digital image processing and artificial neural network. The image processing equipment used in this research was shown in Figure 1.

![Figure 1. Image processing devices for rapid testing of aflatoxins](image)

The equipment consisted of a digital camera (GoPro-4), image capture box, 5 watt UV lamps (4 pieces), and a set of computers. The distance of the camera to the object was 20 cm. The corn grain was placed on the black sample holder and a camera was placed perpendicular to the corn grain with UV lighting. After having acquired image, the image processing was performed to obtain the parameters. All of the parameters were then trained to obtain optimal weights. This training was done many times and stopped after reaching the optimum target.

2.1 Image Processing
Image processing started with thresholding process. The purpose of thresholding was to distinguish the objects with the background of image. Upon thresholding process, the value of parameters was calculated including R, G, B, color value, area, circumference, midline of diameter, hue, saturation and intensity of each pixel of image, either the image for aflatoxin at different concentrations (1 – 2 ppb, 2 – 3 ppb, 3 – 4 ppb, and 4 – 5 ppb).
2.1.1 Parameter measurement RGB (Red, Green and Blue).
RGB parameters derived from each pixel in the color image of corn which is the intensity value for each red, green, and blue. The average value of R, G and B were summed to obtain the color value.

2.1.2 Measurement of parameters area, circumference and diameter midline of aflatoxins spot area.
Measurement of area, circumference and length of each corn was done by changing the image into black and white. Measurements of the area of objects were calculated by counting the number of white pixels. Based on the measurements, the value area of each object was obtained. Pixel tracking procedure which restricts the object with the background done by comparing the color of the object with the background pixels. White pixels bordering the black pixels were the outer most pixels of the object, so that the circumference can be calculated from the sum of the outer pixels. The length of object is obtained by measurements of the distance of each pixel to outer most pixel of the object. The distance values were then compared to find the longest one. To determine the length of the object using Euclidian methods. The distance was obtained by multiplying the number of pixels with a pixel size. The formula used to measure the length was:

\[ d([i_1,j_1],[i_2,j_2]) = \sqrt{(i_1 - i_2)^2 + (j_1 - j_2)^2} \]  

(1)

2.1.3 Parameter calculation of hue, saturation and intensity.
Parameter values HSI (Hue, Saturation, Intensity) was calculated by equation (4), (5) and (6). Intensity was calculated by adding together the intensity of red, green, and blue (RGB) of each pixel in order to obtain an algorithm of gray image.

\[ I = \frac{R + G + B}{3} \]  

(2)

\[ \cos H = \frac{2R - G - B}{2\sqrt{(R-G)^2 + (R-B)(G-B)}} \]  

(3)

\[ S = 1 - \frac{3}{R+G+B} \min(R,G,B) \]  

(4)

2.2 Preparation of ANN model
The ANN architecture that was built consisting three layers, such as input layer, hidden layer and output layer. As an input layer consists of 10 units, those were intensity of red (R), green(G), blue(B), the Color Value, area, circumference, length, and HSI. The output layer consists of 4 units, ie, image of aflatoxins 1 – 2 ppb, 2 – 3 ppb, 3 – 4 ppb and 4 – 5 ppb. While the number of nodes in the hidden layer was as much as (2*n) = 20 nodes. The algorithm used in the artificial neural network was back propagation algorithm with learning rate of 0.3 and 0.5 Logistic Constant.

According to Richand Knight [13], the back propagation training algorithm was as follows:

a. Initialization
- Normalization of the input data \( x_i \) and target data \( t_i \) in the range (0,1)
- The entire weighting \( (w_i) \) given initial random value between -1.1
- Initialization of activation thresholding units, \( x_0=1 \) and \( h_0=1 \)
b. Activation units from input layer to hidden layer using the function:

\[ h_j = \frac{1}{1 + e^{-\sum w_{ij}x_i}} \]  

where:
\( w_{ij} \) = weight that connects nodes \( i \)th in the input layer with nodes \( j \)th in the hidden layer

c. Activation of the units from hidden layer to output layer using the function:

\[ y_k = \frac{1}{1 + e^{-\sum v_{jk}h_j}} \]  

where:
\( \sigma \) = logistic constant
\( v_{jk} \) = weight \( v \) that connects node \( j \)th unit in the hidden layer with node \( k \)th in the output layer

d. Calculating error of the units in the output layer (\( \delta_k \)) and customize it with weights \( v_{jk} \)

\[ \delta_k = (1 - y_k)(t_k - y_k) \]  

where:
\( t_k \) = the target output of the node \( k \)th

\[ v_{jk} = v_{jk\ old} + (\beta \delta_k h_j) \]  

where:
\( \beta \) = constant of learning rate
\( v_{jk\ old} \) = weight previous \( v_{jk} \)

e. Calculating error in the hidden layer (\( \tau_j \)) and customize it with weights \( w_{ij} \)

\[ \tau_j = h_j(1 - h_j)\sum k\delta_k v_{jk} \]  

\[ w_{ij} = w_{ij\ old} + \beta \tau_j x_i \]  

g. Iteration

The whole process was carried out on each sample of each iteration until the system reaches the optimum condition. Iteration involves giving samples of input and output, calculation of activation and change in weight value.

\[ RMSE = \sqrt{\frac{\sum (p_i - a_i)^2}{n}} \]  

\[ Error(\%) = \frac{\sum p_i - a_i}{n} \times 100\% \]
2.3. Validation of ANN Models

Validation was performed as a network performance testing on samples that have not been given during the training process. Network performance can be assessed based on the value of RMSE (Root Mean Square Error) on the generalization of the input-output data, the value of RMSE can be denoted as:

\[
RMSE = \sqrt{\frac{1}{n} \sum_{i=1}^{n} (p - a)^2}
\]

where:
- \( p \) = predicted value generated by the ANN
- \( a \) = the target value which given on ANN
- \( n \) = number of data in the data set of validation

Process of validation was done by inserting the value of data set of input-output which given during the training process. If the ANN has been successful during the training and validation process then the system can already be used for further applications.

3. Results and Discussion

3.1 Generating data of image characteristic of aflatoxin

In order to generate the image characteristic of aflatoxin, the size of image of 640 x 480 pixels or VGA was required. On the preliminary research, 10 parameters of image input were used such as index R, index G, index B, Color Value, area, circumference, midline of diameter, Hue, Saturation and Intensity toward 4 targets such as 1 – 2 ppb, 2 – 3 ppb, 3 – 4 ppb and 4 – 5 ppb. The various images of aflatoxins at different concentrations were shown in Figure 1-5.

![Figure 2. Aflatoxin image 1 – 2 ppb](image)

![Figure 3. Aflatoxin image 2 – 3 ppb](image)
3.2 Image Characteristic
Each characteristic of image was plotted in the graph, to show the value difference of each targets (Figure 6). The differences in the pixels value, gave the information on the most important factors that influenced the recognized the targets.

![Figure 6. Image characteristic of aflatoxin](attachment:image_characteristic.png)

The pictures showed that the characteristics of image could easily be distinguished by area, circumference, and midline of diameter. The exact recognition of image characteristic could be determined by its value of weight.
3.3 Training
Training for each criterion on the content of aflatoxin on using 136 sets of data showed that the accuracy of 100%, demonstrating that the system could recognize the objects accurately. The weight which was obtained from the training can be inserted into the system and then the system could work well. This accuracy value was constant after iterations 1643 times as shown in Table 1.

| Estimated expert | Model estimated | accuracy (%) |
|------------------|-----------------|--------------|
|                  | 4 – 5 ppb       | 3 – 4 ppb    | 2 – 3 ppb | 1 – 2 ppb | Error | Unidentified |
| 4 – 5 ppb        | 39              | 0            | 0         | 0         | 0     | 0           | 100 |
| 3 – 4 ppb        | 0               | 35           | 0         | 0         | 0     | 0           | 100 |
| 2 – 3 ppb        | 0               | 0            | 39        | 0         | 0     | 0           | 100 |
| 1 – 2 ppb        | 0               | 0            | 0         | 18        | 0     | 0           | 100 |

3.4 Validation
In order to determine the accuracy of ANN model, the validation was conducted with 165 sets of data from different samples. The validation results are shown in Table 2.

| Categories    | Samples | Correct Estimation | False Estimation | Unrecognized | Error | Accuracy (%) |
|---------------|---------|--------------------|------------------|--------------|-------|--------------|
| 4 – 5 ppb     | 78      | 78                 | 0                | 0            | 0     | 100          |
| 3 – 4 ppb     | 121     | 121                | 0                | 0            | 0     | 100          |
| 2 – 3 ppb     | 115     | 111                | 4                | 0            | 0     | 97           |
| 1 – 2 ppb     | 62      | 62                 | 0                | 0            | 0     | 100          |

Table 2 above showed that the result of validation for all targets had a very good accuracy value, ie. 100%, except target of 2 – 3 ppb (97%). The average accuracy of this validation was 99%. Improving in the accuracy of the 2 – 3 ppb of corn could be done by recheck the corn increasing the number of sample on training process, so that it would increase the values of accuracy. Based on this validation test the weight of ANN model could be put inside the application program for rapid testing of aflatoxins.

3.5 Application Program for Rapid Testing of Aflatoxin
Application program for rapid testing of aflatoxin on agricultural products can be easily operated by anyone. The application program was made by using computer programming and it can be used interactively by user. The interface of rapid testing program of aflatoxin is shown in Figure 7.

Figure 7. Application program for rapid testing of aflatoxin

Application program for rapid testing of aflatoxin was operated by using Microsoft Windows. The application program can be operated directly by clicking the button that available in monitor.
Procedure operating the application program is as follows: (1). Prepare the sample box and plug in the electricity devices, (2). Make sure that the UV-light is adequate inside the sample box, (3). Put camera on sample box and make sure that the camera’s lens is located right on the shooting hole, (4). Prepare the material to be tested, (5). Make sure the camera is focused on the image, (6). Capture picture by clicking the button on the camera, (7). Change the image resolution to VGA, (8). Call the image by clicking on the "BUKA" button in the application program, (9). Click the "PROSES" button, then the program will automatically work to do the processing and directly display the results, (10). If we will do the test again, then first click the "REFRESH" and "DELETE", (11). Click the “EXIT” button to finish and axit.

4. Conclusions

Digital image processing has been used to generate the numerical data of the physical characteristics of aflatoxin image such as value of RGB, HSI and physical characteristic such as area, circumference and diameter midline of aflatoxins, they have a specific value. The most influencing input factor to recognize the object was the diameter midline of aflatoxins. This factor can be used for developing the rapid testing of aflatoxins devices.

Application program for rapid testing of aflatoxin by using image processing and artificial neural network has the ability to recognize the aflatoxin contamination in corn 100% with validation of 99%. This application can be operated by computer equipped with a sample box for acquisition image. The device of rapid testing aflatoxin can be operated easily by the user.

5. References

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