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Prediction of Fermentation Index and pH of Cocoa (*Theobroma cacao* L.) Beans Based on Color Features (Cut Test) and Partial Least Square Regression Model

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Abstract Post-harvest processes such as the fermentation stage are important parameters to determine the quality and price of cocoa beans. The color change of cocoa beans from dark purple to brown is an indicator of success in the cocoa fermentation process. So far, farmers use an estimation system and use laboratory equipment which has many shortcomings and is difficult to apply to farmer groups. The application of technology to measure the rate of fermentation quickly and accurately encourages several scientists to build a predictive model that is used as the initial stage of making a technology. The purpose of this study was to determine the prediction results of the Fermentation Index and pH of cocoa beans using a partial least square regression model and to determine the relationship between the image of cocoa beans (cutting test) with the pH and fermentation index of cocoa beans. The method uses primary data from research in the laboratory for pH and cocoa fermentation index measurements and cocoa bean image processing to develop a predictive model. The result of this study is a partial least square regression model for predicting the cocoa bean fermentation index with cross validation 5 times shown that, partial least square regression model for pH prediction shows an accuracy of 99% and the error value is only 0.007.

Keywords Color Analysis, Image Processing, Partial Least Square Regression

1. Introduction

Cocoa is a tropical plant that is widely bred in Indonesia with the 3rd largest production in the world in 2010 with an increase in production of 6.63 percent per year [1]. Fermentation is an important parameter to show the quality of cocoa beans as an export commodity. Improvement of the fermentation process is an important parameter to determine the quality and price of cocoa beans in order to compete in the world market. This improvement effort aims to obtain cocoa beans with perfect fermented standards and water content below 7.5% [2].

The importance of the cocoa fermentation process is because during the fermentation process there is a decrease in the pH of the cocoa beans and inactivation of enzymes that produce flavor precursors [3].

The enzyme is able to hydrolyze pectin which causes the pulp tissue to decompose, forming a liquid and dripping

out of the seed pile. On the other hand, microbial activity in the decomposition of pulp sugar causes formation of organic acids that cause high levels of acidity in dry cocoa beans [4], [5]. The reaction between sucrose and protein will produce a chocolate flavor precursor which is influenced by the physical properties of the cocoa bean. The physical properties of cocoa beans in the form of a darker purple color change require a longer time than bright beans [2]. An indicator of the success of the cocoa fermentation process occurs when the color change from unfermented cocoa beans is dark gray (gray) to dark purple and brown in completely fermented cocoa beans [6].

Currently, the cocoa fermentation process among farmers uses wooden boxes which have drawbacks in the form of not being able to control both temperature, pH and fermentation index. So far, cocoa farmers have only used an estimation system or determining the fermentation degree to determine the level of fermentation quality from cocoa beans even though this method is subjective and inaccurate and requires a long testing time [7]. This fermentation process produces cocoa beans of low fermentation index (grades 3 and 4). Around 85% of the national production of cocoa beans is not fermented. It has an impact on the selling price of cocoa beans in the international market which gets a price reduction of 10-15% from the market price [8]. The importance of applying technology to measure fermentation quickly and accurately to measure quality control of cocoa beans has prompted several scientists to build a predictive model that is used as the initial stage of making a technology.

Partial Least Squares Regression is a regression model to solve multicollinearity problems in multiple linear regression without reducing the variables included in the model that is suitable for predicting the fermentation rate of cocoa beans. This PLSR is indirect where the variance of the dependent variable is related to the independent variable through one or several factors which are defined as a linear combination of the independent variables called components. PLSR is a multiple linear regression which has many advantages such as being able to solve the problem of small research sample sizes (microarray data), missing data (missing values), multicollinearity occurs, and can be applied to different data types. While the drawback of this PLSR is the great difficulty in interpreting data with a number of latent independent variables (which is based on the cross-product relationship with the response variable, unlike factor analysis based on the covariance matrix between the explanatory independent variables for latent variables) [9,10]. Based on this research, this article aims to develop a prediction model of Fermentation Index and pH of cocoa beans using a partial least square regression model based in digital image features and additionally to provide insight on the fermentation index and pH change during fermentation process of the samples.

2. Materials and Methods

The research was conducted from August to December 2021. The research was conducted using a mini studio and *cutting test* located in the Faculty of Agricultural Technology, Universitas Brawijaya and the research stage at the Laboratory of Pharmaceutical Chemistry, Faculty of Pharmacy, Ma Chung University for the measurement of the fermentation index. The sample of cocoa beans used came from Indonesian Coffee and Cocoa Research with Trinitario varieties. The samples of cocoa beans analyzed were 150 samples from 10 categories. The cocoa image segmentation process uses the Otsu method form of a histogram to group pixels on the x and y axis images. The x-axis in the Otsu method represents a different level of intensity, while the y-axis represents the number of pixels that have that intensity value [11]. The data analysis phase uses Python 3.6 software with scikit learn library. The image acquisition process is carried out by taking image data using LED lighting (mini studio) and a color chart as a reference. The mini studio and *cutting test* used look like the picture below.

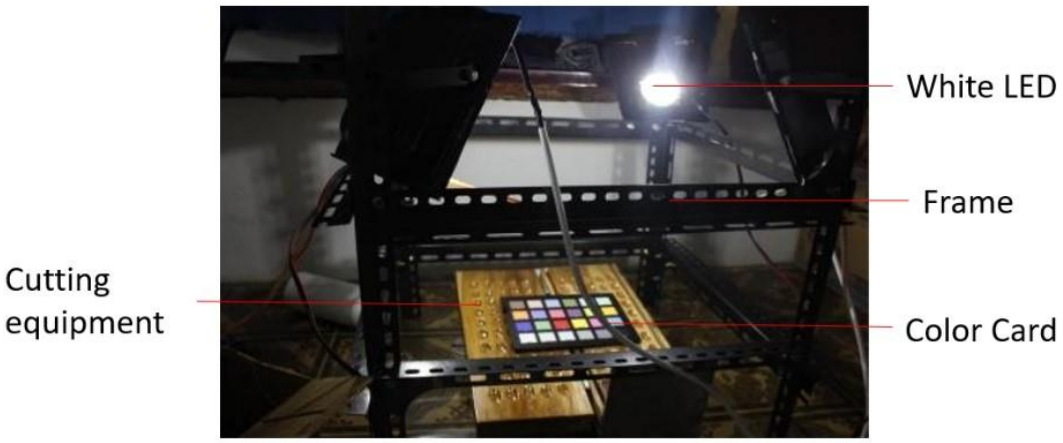


Figure 1. Mini Studio

2.1. Cleavage of Samples of Cocoa Beans

Samples of dried cocoa beans with a moisture content of <7.5% were split using a *cutting test* with a capacity of 50 cacao beans for each cleavage process. Figure 2 shows the split cocoa beans.

2.2. Image Acquisition

The process of taking pictures of cocoa beans from each category was carried out in a modified mini studio with a size of 50 x 50 x 60 cm and a light intensity of 1394 lx. The EOS Utility application is used to be able to adjust the ISO, *exposure*, and contrast of the resulting cocoa bean image. Setting the image at ISO 6400, *shutter speed* 1/30, Aperture F/2.5 and setting the camera focus by rotating the lens on the front of the camera.

2.3. The Measurement of Fermentation Index from Cocoa Beans

The Gourieva and Tsernetivinov method was carried out with 0.5 g of crushed cocoa beans extracted using 50 ml of a mixed solution of methanol: HCl (97: 3). The sample mixture with the solution was then left homogeneously in the refrigerator (80 ℃) for 20 hours. The filtrate was then filtered using Whatmann paper No. 1. The absorption spectrum was observed at a wavelength of 400–700 nm using a UV-vis Shimadzu UV-1601 spectrophotometer. The fermentation index is calculated based on the ratio of the absorbance value at a wavelength of 460 nm to an absorbance of 530 nm [12-14].

2.4. The Measurement of pH from Cocoa Beans

Measurement of the pH of seed chips using a pH meter. The sample of cocoa beans taken is a sample of beans that have been imaged with a total of 5 beans for each fermentation category. The method for measuring pH is as follows: Samples of cocoa beans are taken from 3 points, namely the top, middle and bottom points. A sample of cocoa beans was ground and then 1 gram of cocoa beans was weighed to measure the pH. Dissolved in 5 ml of distilled water, then stirred with a glass stirrer for 3 minutes. The pH was measured using a pH meter [12,14].

2.5. Image Analysis

Feature extraction stage on cocoa beans using RGB, Lab and HSV color features as input dataset from the prediction model and output dataset value from laboratory test results for pH and fermentation index. Destructive dataset processing is carried out using a Jupyter Notebook Type Python 3.6 using the scikit-learn library using the coding in Figure 3 for the PLS regression algorithm. The model used for the analysis of the 50 extracted data is partial least squares regression. Partial Least Squares Regression is used to calculate R^2 and MAE and evaluate the resulting model. The PLS model consists of a predicted pH and fermentation index.

Figure 2. Samples of Cocoa Beans (Cut Test)

3. Result and Discussion

3.1. Measurement of pH from Cocoa Beans

The results of measuring the pH of cocoa beans using a pH meter greatly affect the taste and aroma characteristics of the cocoa products produced. Cocoa beans with a pH of 5.0–5.5 produce a higher aroma potential when compared to a pH of 4.0–4.5 which gives a lower aroma [15]. Dry cocoa beans without fermentation before incubation usually have a pH of 6.12 and experience a decrease in pH after incubation. Samples of cocoa beans with a total of 5 samples for each level of fermentation of cocoa beans are shown in Figure 3 with *standard deviation* for each time.

During the fermentation process, the cocoa beans experienced a decrease in pH on the 1st and 2nd day of fermentation. Cocoa beans will increase slightly on the third day until the last day of fermentation. The decrease in pH in the first 2 days of fermentation was caused by the cocoa bean pulp which contains citric acid which allows the growth of bacteria. However, as the fermentation progresses, the bacteria (yeast) that dominate the good pectinolytic activity will degrade the cocoa pulp and release citric acid, allowing the growth of other bacteria. This also causes an increase in the pH of the pile of cocoa beans on the 5th day. Changes in the pH of the heap of cocoa beans were caused because during the fermentation process air entered the pile of cocoa beans accompanied by the entry of bacteria, namely acetic acid bacteria, resulting in acid penetration of the cocoa beans [16]. The decrease in pH value was caused by the diffusion of acetic acid into the cocoa bean layer, resulting in an acidification process. Acid that diffuses into the cell wall will cause the death of cocoa bean cells [17]. Powdered cocoa bean samples with incubation resulted in the lowest pH values compared to other treatments due to differences in the rate of acid diffusion into the cotyledons. The surface area of cocoa beans in contact with acetic acid greatly affects the speed of penetration of acetic acid. Incubation stage and after drying, the pH value of the cocoa bean powder treatment increased when compared to other treatments. This happens because during the drying process volatile acid compounds evaporate along with the evaporation of water. In addition, during drying, air from the outside will enter through the seed coat so that the cell fluid contained under the seed coat evaporates and slowly the cotyledons will turn brown. Browning is an oxidation reaction of polyphenolic compounds with the help of air and polyphenol oxidase [4,18].

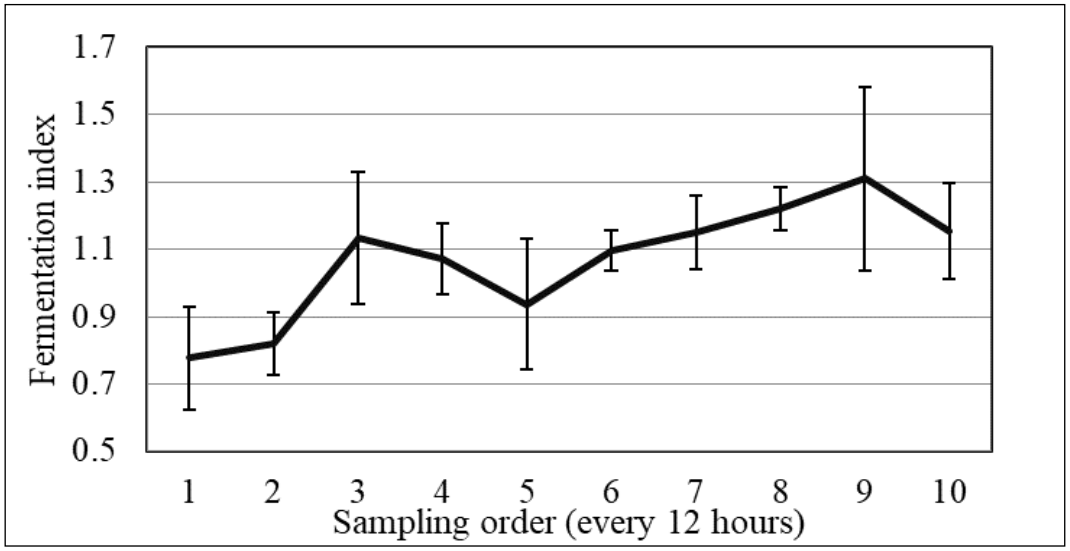


Figure 3. pH of Cocoa Bean

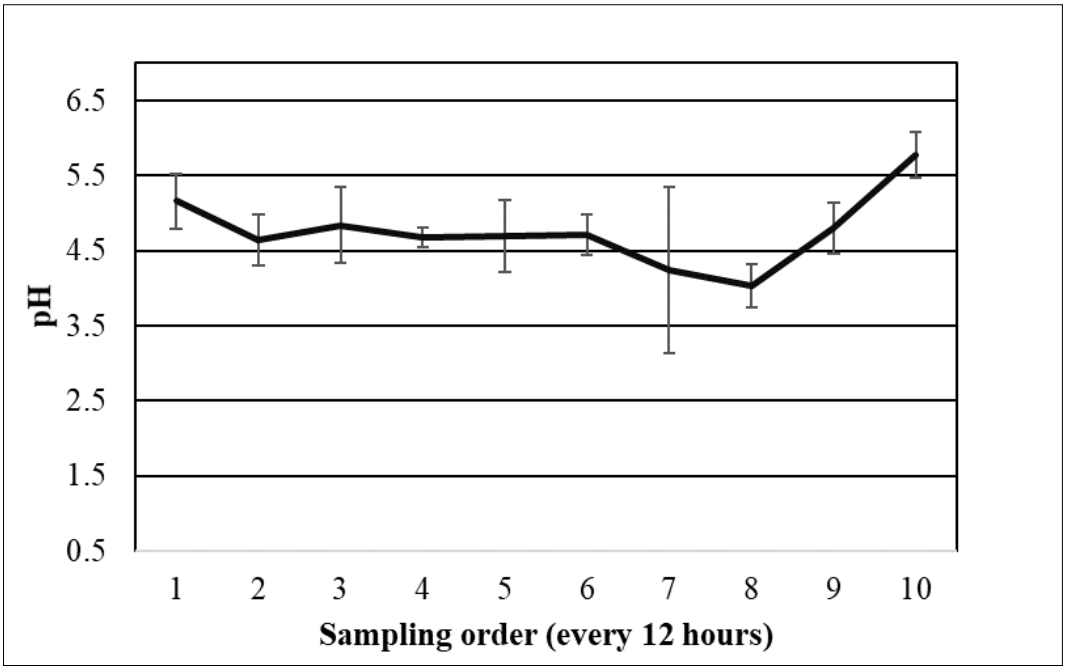


Figure 4. Fermentation Index of Cocoa Bean

3.2. Measurement of Fermentation Index of Cocoa Beans

The results of the measurement of the fermentation index of 5 samples for each level of fermentation of cocoa beans that have been imaged and pH taken using the Gourieva and Tsernetivinov method are shown in Figure 4. The results of the measurement of the fermentation index using the 460 and 530 nd waves of each sample.

Fermentation index of each sample increased with the addition of the cocoa bean fermentation time. The results of the measurement of the cocoa bean fermentation index showed that there was an increase in the fermentation index from the 1st time to the 3rd time. And there was a decrease from the 4th time to the 5th time, but it increased to obtain a fermentation index of 1 at the 10th time. This happens that the longer the fermentation time, the fermentation index of cocoa beans also increases until it reaches a value of 1 which indicates perfectly fermented [19]. While the results of the measurement of the level of fermentation of cocoa beans in the sample have a fermentation index that is fluctuating from category 1 to 10. Fluctuating fermentation index values can occur due to several factors of cocoa bean variety and initial conditions of fermented cocoa beans, dimensions of the fermentation container, degree of aeration the container, the thickness of the pile of beans in the fermentation container, and the length of the fermentation process [20]. In addition, the surface area of cocoa beans and the stirring of cocoa beans have a significant effect on the value of the cocoa bean fermentation index, this is one factor in the fluctuating value of the cocoa bean fermentation index [14].

3.3. Image Acquisition and Dataset Building

Cocoa beans with a total of 5 samples in each category were analyzed for the relationship between pH and color features and the relationship between fermentation index and color features. The picture of each category of fermentation has been shown in Figure 5.

The dataset was obtained by performing several stages using a python application such as data loading, segmentation using the Otsu method, *cleaning* and color feature extraction. The dataset which has been combined with the results of the physicochemical test of pH and fermentation index is obtained by using the *Partial Least Squares Regression Model.* Table 1 shows dataset color images of cocoa beans that have been extracted.

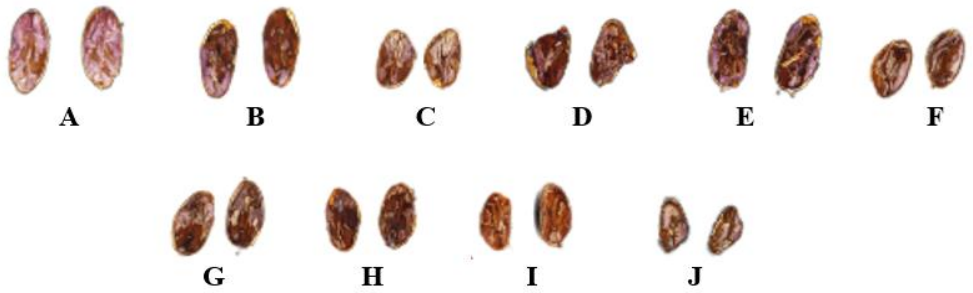


Figure 5. Samples of Cocoa Beans from Fermentation Level Category 1-10

Table 1. Cocoa Color Image Feature and Prediction Target Dataset

R_ave	G_ave	B_ave	L_ave	a_ave	b_ave	H_ave	S_ave	V_ave	pH (Y1)	IF (Y2)
58.15	36.36	27.83	39.34	108.42	89.91	39.34	108.42	89.91	5.3	0.63
39.59	28.82	26.30	30.74	84.87	75.62	30.74	84.87	75.62	5.6	0.76
41.66	29.94	26.81	31.79	92.74	82.56	31.79	92.74	82.56	5.1	0.71
….	….	….	….	….	….	….	….	….	…	…
….	….	….	….	….	….	….	….	….	…	…
34.81	24.34	21.49	25.05	88.71	79.60	25.05	88.71	79.60	5.7	0.95
44.29	28.41	22.97	29.7	101.05	87.35	29.7	101.05	87.35	6.1	1.34
25.29	20.02	19.12	20.04	77.41	72.76	20.04	77.41	72.76	6.1	1.21

ave = average

3.4. Partial Least Square Regression Analysis

The results of making the PLS model are suitable for use on a label that has a value *such* as the pH value and the cocoa bean fermentation index of each sample. From the results of the relationship between color features and the fermentation index using the division of 0.20 *testing* and 0.80 *training* with a random value of 42 and using the method, *cross validation* 5 times with random values of 1 and 3 components as shown in Table 2.

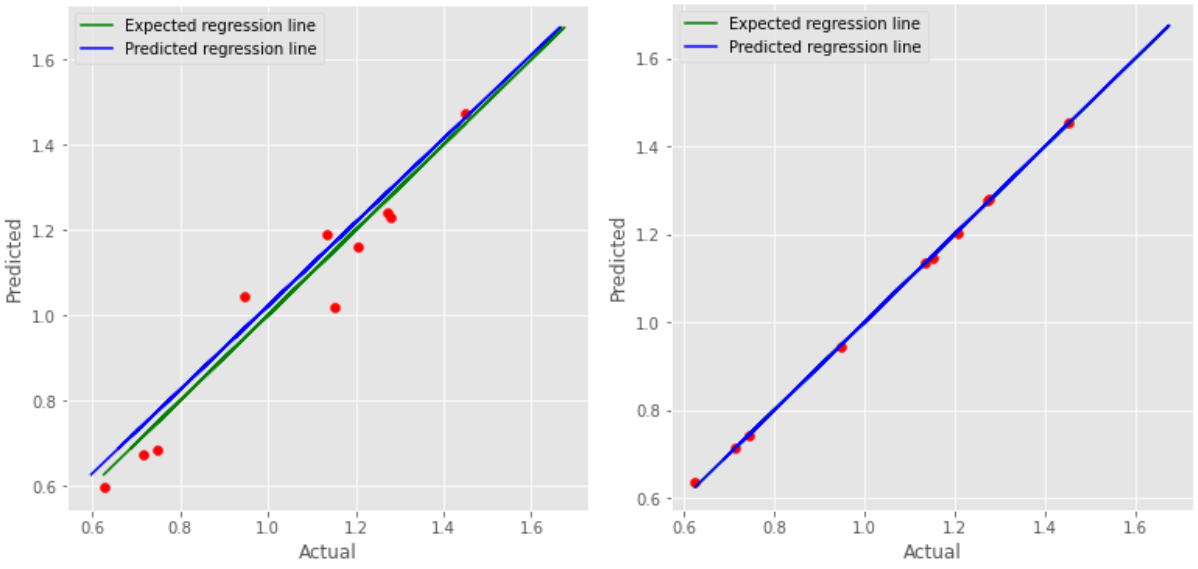
Table 2. PLS Regression from Fermentation Index

	R^2 data *training*	R^2 data *testing*	MSE *training*	MSE *testing*
PLS	0.916	0.939	0.057	0.065
PLS dengan CV	1.0	0.99	0.0036	0.0047

Based on the results of the analysis, it can be seen that there is a significant correlation between the value of the X variable in the form of color features and the cocoa bean fermentation index as seen from the high R^2 with low error value. The value of the cocoa bean fermentation index is strongly influenced by the length of the fermentation time and the curing process of the cocoa beans which have a significant effect on the cocoa bean fermentation index [21]. Selection of the approach using RGB color features, Lab and HSV provides benefits as a visual approach to detect the level of fermentation of cocoa beans by matching the value of the cocoa bean fermentation index [3]. The correlation plot of the predicted and actual values of the PLS method used can be seen in Figure 6.

The results of the analysis of the relationship between pH and the results of color feature extraction using the PLS regression method were carried out with a ratio of 25:75 for testing and *train* with a random value of 42 and the use of PLS regression *cross validation* 5 times with random values of 1 and 3 components which resulted in accuracy. and errors that can be seen in Table 3.

This shows that there is a significant relationship between pH and the color features of cocoa beans with a high R2 value from the *training* and *testing* which shows a significant relationship. Good quality fermented cocoa beans have a pH between 5.2–5.5 which can produce a distinctive cocoa aroma [21,22]. The results of the correlation of pH with color features with the PLS method can be seen in Figure 7.

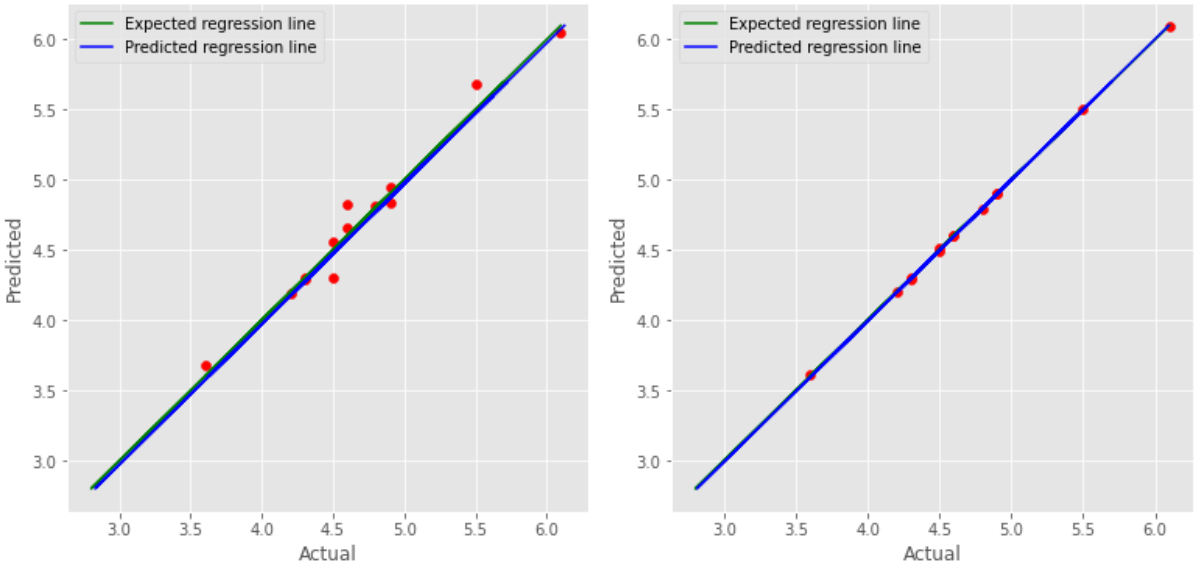


(A) (B)

Figure 6. Correlation Results of Fermentation Index with Color Features (A) PLS without modification, (B) PLS with *cross validation*

Table 3. PLS Regression of pH

	R^2 data *training*	**R^2 data *testing***	**MSE of *training***	**MSE of *testing***
PLS	0.97	0.96	0,1060	0,1061
***PLS* dengan CV**	0.99	0.99	0.0108	0.007



(A) (B)

Figure 7. Correlation of pH with Cocoa Bean Color Features (a) PLS Regression Without Modification (b) PLS Regression with *cross validation*

4. Conclusions

Based on the results of the analysis of 10 categories of cocoa bean fermentation level, it shows that if the prediction results of the fermentation index use the partial least square regression model with the *cross validation* 5 times, using a division of 20:80 shows that it has the highest level of accuracy, which is 99% for testing data and the error value is only 0.0047. In the fermentation pH prediction model using the partial least square regression model with the *cross validation* 5 times, the data distribution of 25:75 shows an accuracy of 99% for testing and training data with an error value of only 0.007 for testing data. In addition, based on the calculation of pH and cocoa fermentation index quantitatively, it shows that controlling temperature, container area, surface area of cocoa beans and stirring affect the diversity of fermentation rates of each cocoa bean.

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