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Research Article

Estimation of single leaf chlorophyll content in sugar beet using machine vision

Parviz Ahmadi MOGHADDAM[1,*], Mohammadali Haddad DERAFSHI[1], Vine SHIRZAD[2]
[1]Department of Agricultural Machinery, Agricultural Faculty, Urmia University, Urmia - IRAN
[2]Department of Plant Protection, Agricultural Faculty, Urmia University, Urmia - IRAN

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Abstract: Estimating crop nitrogen status accurately during side-dressing operations is essential for effective management of site-specific nitrogen applications. Variable rate technology (VRT) is one of the major operations in precision agriculture to reduce environmental risks and increase fertilizer use efficiency. In the present study, color image analysis was performed to estimate sugar beet leaf chlorophyll status. The experiment was carried out in a phytotron and nitrogen was applied at 6 levels to the sugar beet grown in pots. Chlorophyll level of the leaves was measured by a SPAD-502 chlorophyll meter. To estimate chlorophyll status, a neural-network model was developed based on the RGB (red, green, and blue) components of the color image captured with a conventional digital camera. The results showed that the neural network model is capable of estimating the sugar beet leaf chlorophyll with a reasonable accuracy. The coefficient of determination (R2) and mean square error (MSE) between the estimated and the measured SPAD values, which were obtained from validation tests, appeared to be 0.94 and 0.006, respectively.

Key words: Machine vision, neural network, chlorophyll, sugar beet, variable rate

Introduction

Nitrogen (N) is a nutrient critical to the growth of agricultural crops. Proper management of nitrogen application helps to reduce nitrogen losses and prevents pollution of underground and surface water, which leads to serious environmental problems (Noh et al. 2006). Traditionally, fertilizer is applied onto the whole farmland regardless of the variations across the land (Sindir and Tekin 2002). Compared with conventional nitrogen application methods, variable rate technology (VRT) has less potential for nitrogen leaching. Moreover, VRT is economically competitive (Roberts 2008; Biermacher et al. 2009).

The effectiveness of variable rate of nitrogen application heavily relies on the capability of detecting nitrogen status while the fertilizer is being applied (Noh et al. 2006). There are several methods for detecting plant nitrogen content including plant analysis, leaf chlorophyll measurement, and remote sensing systems. Although soil and plant analyses are very precise methods to obtain nitrogen status, they are very time consuming and expensive approaches and not appropriate for variable rate nitrogen application (Noh et al. 2006). Leaf chlorophyll measurement is a very good criterion for estimating crop nitrogen status (Gitelson and Merzlyak 1997;

* E-mail: p.ahmadi@mail.urmia.ac.ir

Shapiro 1999). Nevertheless, it has limited potential for N management in large fields due to the large number of chlorophyll samplings that are required for precise nitrogen status mapping (Miao 2009). This limited number of data points may inadequately represent the distribution of crop nitrogen stress across the field.

To obtain more complete and accurate information on nitrogen status for increasing the effectiveness of nitrogen management, researchers developed various methods to estimate leaf chlorophyll content based on spectral images of the crop canopy (Liangliang et al. 2004; Namrata et al. 2007). Digital processing of images has been used with success in crop management and detection of nitrogen stress (White et al. 2000; Koumpouros et al. 2004; Pydipati et al. 2006). The development of low-cost digital cameras that use charge-coupled device (CCD) arrays enabled the capture of images for estimating plant nitrogen status. This technology was used for the first time by Woebbecke et al. (1995) to split images into different areas, representing soil and plants. Furthermore, Liangliang et al. (2004) and Pagola et al. (2009) successfully developed a conventional digital camera for assessing chlorophyll content of winter wheat and barley, respectively.

Selection of an appropriate technique in obtaining a robust prediction model to be used for leaf chlorophyll content is a critical task. Artificial neural networks with error back-propagating training (BP-ANN) have been widely applied to non-linear models (Chen et al. 2007). Chen et al. (2007) developed an ANN model for predicting the chlorophyll content of cabbage seedlings. Their results indicated that the ANN model could be used to develop a practical remote sensing system with reasonable accuracy (R^2 = 0.93).

The present research developed models to estimate sugar beet leaf chlorophyll, based on processing of color images captured with a conventional digital camera during variable rate operations. In the present work, 8 regression models and 1 neural network model are presented. These were compared with the regression model proposed by Pagola et al. (2009).

Materials and methods

This experiment was carried out in October 2008, in a phytotron in Urmia University, Urmia, Iran. The sugar beet seeds were planted in 30 plastic pots of 22 cm diameter and 25 cm height filled with a loamy soil. The air temperature and the relative humidity of the chamber were adjusted to 25 and 20 °C and 50% and 60%, for day and night, respectively. Nitrogen was applied as urea (46% N) at 6 levels (0, 100, 200, 300, 400, and 500 kg ha^{-1}) and each treatment was replicated 5 times. Thirty days after sowing through the end of the experiment, at 5-day intervals, 6 leaves, as representatives of each pot, were separated from the plants. Thus, 180 images were acquired in this experiment. Images were taken from each sampled leaf by a digital camera (DSC W200, Sony, Japan) installed at 50 cm height and simultaneously chlorophyll concentrations of leaves were measured using a chlorophyll meter (model SPAD-502, Minolta Co, Japan). To achieve reasonable images without reflection and also reduce the noise, different backgrounds were tested. The black fabric was good for this purpose (Figure 1). The images were recorded in jpeg format with the resolution 2048 × 1536 pixels. Digital color images consist of 3 components, red (R), green (G), and blue (B), and each component had 256 graduations. MATLAB software (Version 7.6, 2009, Mathworks Company) was used for image processing.

Machine vision

The machine vision was carried out by separating the original image of individual leaf into 3 monochrome images including red, green, and blue. The reflectance of the sugar beet leaf was stronger than its background in monochrome images (Figure 2). Therefore, the following threshold function was employed to segment the leaf from the background:

$$g_i(x,y) = \begin{cases} 0 & f_i(x,y) \le T_i \\ f_i(x,y) & f_i(x,y) \ge T_i \end{cases} \quad (1)$$

where $g_i(x,y)$ is the segmented gray level at pixel (x, y), $f_i(x,y)$ is the original gray level at pixel (x, y), T_i is a threshold value, and i represents the red, green, and blue components. Threshold values were calculated for the 3 components by training-error method and automatic thresholding.

Figure 1. Sugar beet single leaf with background.

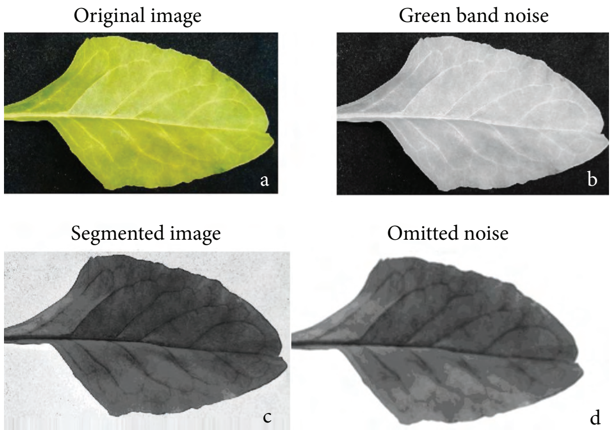

Figure 2. Steps of image processing a) original image b) green band image c) leaf segmented from background d) noise omitted.

As Eq. (1) shows, the pixel values of the leaf are not changed and the pixel values of the background equal zero. In the next step, the sugar beet leaf reflectance was calculated on the basis of average intensity values of the monochrome images. The average intensity values were calculated as:

$$A_i = \frac{\sum g_i(x,y)}{N_i} \qquad (2)$$

where A_i is the average intensity value at component *i*, $g_i(x, y)$ is the intensity value of the pixel representing sugar beet leaves, N_i is the total number of pixels representing sugar beet leaves in component *i*, and *i* represents the R, G, or B components of the image.

Neural-network model development

In the present study, a multilayer perception neural network (MLPNN) with a back propagation learning algorithm was selected to develop the model for predicting leaf chlorophyll content. Typical MLPNN architecture has 1 input layer, 1 or more hidden layer(s), and 1 output layer. In our study, the neural-network model was trained using the reflectance of sugar beet leaves detected at R, G, and B components as the inputs and the corresponding measured SPAD readings as the output. Their input and output, together, develop a matrix for training neural network model as shown in Eq. 3:

$$\text{MT} \ \ [\text{red, green, blue, SPAD}] \qquad (3)$$

This information was used as the training data sets for the proposed neural-network model development.

The topological structure of MLPNN model in this research consisted of 3 neurons in the input layer and 1 neuron in the output layer to match the 3-1 input–output pattern of the training data set. Before the training process, the input–output data pairs were randomly selected from the data set (from 180 data). In the present study, 70% of the data set (126 data) was selected as training data (Demuth and Beale 2000). Predetermined values for the output error (MSE) and maximum iteration number were set to 0.001 and 5000 epoch, respectively. Since the accuracy of estimation is highly dependent on covering all level of data, the randomization process was repeated until a satisfactory level of data distribution was reached. The error back propagation (BP) training algorithm compares the estimated output value obtained from the model with the corresponding measured value. The difference between these values was considered the error value. The error value will be reduced to reach the predefined value by adjusting the weighing values connecting all the neurons. The training process will be completed when all weighing indices are fixed and the neural-network model can accurately estimate the output data as a function of input values (Hassoun 1995). Both the number of hidden layers and the number of neurons in hidden layer were determined by the training error method.

In the present study, mean square error of prediction (MSE), minimum prediction accuracy (MPA), and coefficient of determination (R^2) were considered to evaluate the performance of neural network model and linear regression models. These criteria were calculated using the following equations of 4, 5, and 6:

$$MPA = \min(P_i \ldots\ldots\ldots P_N) \qquad P_i = \left[1 - \frac{|y_{ai} - y_{pi}|}{y_{ai}}\right] \times 100 \tag{4}$$

$$MSE = \frac{\sum_{i=1}^{N} (y_{ai} - y_{pi})^2}{N-1} \tag{5}$$

$$R_2 = \frac{\left(\sum_{i=1}^{N} (y_{ai} - \bar{y}_a)(y_{pi} - \bar{y}_p)\right)}{\sum_{i=1}^{N} (y_{ai} - \bar{y}_a)^2 (y_{pi} - \bar{y}_p)^2} \tag{6}$$

where in the above equations, y_{ai} and y_{pi} are actual and estimated leaf SPAD, respectively, and $\bar{y}_a$ and $\bar{y}_p$ are average of actual and estimated leaf SPAD, respectively.

Moreover, linear regression models were also analyzed and the obtained results were compared with the neural network model. In general, the goal of linear regression is to find a line that best predicts Y (chlorophyll) from X (red, green, and blue components). The most common method for fitting a regression line is the method of least-squares. This method calculates the best-fitting line for the observed data by minimizing the sum of the squares of the vertical deviations from each data point to the line (if a point lies on the fitted line exactly, then its vertical deviation is 0). In order to find the reasonable approach to estimate chlorophyll concentration (SPAD values) of the leaves, different functions of R, G, and B values were used for linear regression analysis.

Method proposed by Pagola et al. (2009) (I_{pag})

Pagola et al. (2009) used a conventional digital camera to analyze the color of barley leaves as a measure of the amount of chlorophyll in a plant. After studying 4 different indices and comparing with the previous model, they found that Eq. 7 yields the best estimate of chlorophyll content in barley leaves using only the data from the conventional digital camera.

$$I_{pog} = 0.76|R-B| - 0.12|R-G| + 0.64| = G-B| \tag{7}$$

Results

The results of the present study showed that the machine vision is a suitable approach for investigation of plant nitrogen status. The results also indicated that the neural network model trained with the RGB component was capable of proper estimation of chlorophyll level of sugar beet leaf ($R^2 = 0.94$). Different number of hidden layers, different number of neurons in each hidden layer, and different transfer functions between layers were examined. The optimum model obtained from this examination consisted of 1 hidden layer with 10 neurons and the sigmoid function in the hidden layer and the linear function in the output layer.

To validate the model, the remaining of data set (30%) was used as the input to the model. Regression analysis of the output values by neural network model resulted in the following regression equation:

$$SPAD_{est} = 1.05\ SPAD_{mes} - 1.16 \tag{8}$$

where $SPAD_{est}$ is the value estimated by the model and $SPAD_{mes}$ is the value measured by the chlorophyll meter.

The MLPNN model, compared with the linear regression model, showed better results in estimating chlorophyll level ($R^2 = 0.94$ versus $R^2 = 0.88$) (Figure 3). MSE and MPA values for MLPNN function, compared with the linear regression model, were also better (0.006 and 71% versus 0.008 and 58%), indicating higher performance for MLPNN (Table 1).

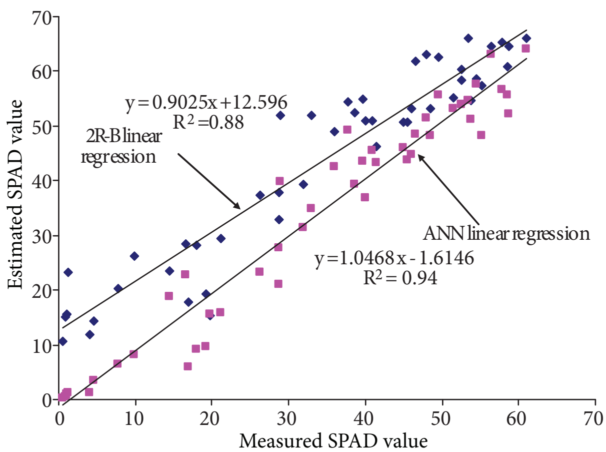

Figure 3. Comparison of MLPNN model and the 'best-fit' regression model, for estimating the SPAD values.

Table 1. Comparison of neural network model and the linear regression model in estimating sugar beet leaf chlorophyll content.

Models	Input variables	MSE	MPA %	Coefficient of determination (R^2)
MLPN	R, G, B	0.006	71	0.94
R, B (regression)	R, B	0.008	58	0.88
$I_{pag}^{(1)}$	R, G, B	-	-	0.91

The model used by Pagola et al. (2009)

Discussion

The study of the obtained results by neural network model indicated remarkably higher accuracy in comparing the estimated and measured chlorophyll levels (MSE = 0.006). Noh et al. (2006) and Gautam and Panigrahi (2007) obtained similar results on corn plants (MSE = 0.03 and MSE = 0.066, respectively).

The models 2R - B and 2R + G - B, among the various linear functions, showed acceptable correlation and resulted in R^2 values of 0.88 and 0.86, respectively (Table 2). Similar results were obtained for wheat by Kawashima and Nakatani (1998), where the (R - B) / (R + B) and R - B models were used (R^2 = 0.81 and 0.76, respectively). This could be attributed to the distinctive difference that existed between the red and the blue components in leaves with higher chlorophyll, compared to lower chlorophyll leaves (Figure 4).

Table 2. Linear regression models in estimating leaf chlorophyll content.

Functions	Equations	R^2 values
R, B	$SPAD_{est} = -0.32(2R - B) + 83.1$	0.88
R, G, B	$SPAD_{est} = -0.221(2R + G - B) + 92$	0.86

The preference for the neural network model can also be attributed to the relationship between RGB components and leaf chlorophyll level, which was not perfectly linear. This was also experienced by Wang et al. (2009) on assessing rape plant nitrogen concentration.

The overall results obtained from the present experiment verified that the MLPNN model can provide more accurate estimations of sugar beet leaf chlorophyll, compared with the linear regression model.

Conclusions

The present study demonstrated that a simple method can be used to estimate the chlorophyll level of sugar beet plant by analyzing the leaf image color. This method, compared to the other remote sensing approaches, is low in cost and easy to perform. The research also developed a neural network model, trained with red, green, and blue components, and

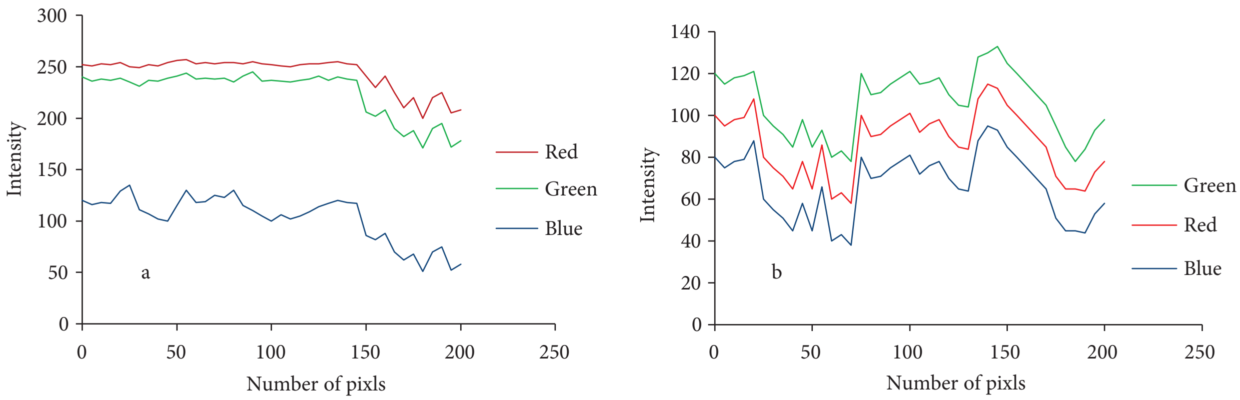

Figure 4. Variation of red, green, and blue component a) leaf with high nitrogen b) leaf with low nitrogen.

compared this model to the linear regression model, which resulted in estimating the chlorophyll level of the sugar beet leaves with higher accuracy. The neural network model developed in the present study performed even better than the model proposed by Pagola et al. (2009), the correlation values being approximately 3% higher.

Since the method of taking image, image processing, and chlorophyll measurement in the present study can be similarly applied to some others plants, similar results might be expected. However, 2 different aspects need to be recommended for use of this method in fields. First, the illumination variation during image taking in fields must be considered. Second, the estimation of plant chlorophyll should be carried out on the plant canopy, where in the existing experiment this assessment was performed only on separated plant leaves.

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