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The Optimization Studies of AB Mix Fertilizers for *Capsicum frutescens* in Soilless Media using Response Surface Methodology Software

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ABSTRACT

The optimization studies of AB mix fertilizers to enhance the growth of *Capsicum frutescens* have been carried out using response surface methodology software, RSM. All plant samples were prepared using wick fertigation technique at different wick lengths: 17, 19 and 21 cm and fertilizers concentrations: 1.8, 2.2 and 2.6 mS/cm. In this study, central composite design of face centered (k = 1) configuration was applied and the quadratic model considers only two parameters i.e.: fertilizers concentrations, X_1 and the wick lengths, X_2. Results showed that X_1 and X_2 gave significant influence on the selected response variables. Statistical analyses explained the coefficient of determination (R^2 < 0.9915), P-value (< 0.0001), F-value (163.54), and not-significant lack of fit values (> 1.14) supporting the validation of prediction models. AB mix fertilizer optimization condition for *Capsicum frutescens* has been identified at 2.54 mS/cm of AB fertilizer and 18.15 cm wick length, with the maximum plant height recorded at 38.73 cm.

1. Introduction

The rapid growth of the world's population has led to an increasing demand for food supply. Many alternatives have been taken to ensure the fresh food supply is secured [1]. *Capsicum frutescens*, (*C. frutescens*), also known as bird's eye chilli is one of the most important crops in Malaysia however, concern about the shortage of chilli supply for local's market demand has been prolonged and therefore, given rise to imported chilli supply from India, China and Thailand to fulfil the local demand [2]. To increase the output with the increasing price of the input is a challenge; an appropriate amount of fertilizers and pesticides are important for food management resources to enhance the quality and quantity of crops. Conversely, excessive amounts and disproportionate use of fertilizers and pesticides can be harmful to the environment and cause serious pollution and health hazards [3]. This includes the compilation of heavy metals in soils that are absorbed and accumulated in fruits, vegetables and grains which, therefore, affects food safety as the bio-concentrated pollutants enter organisms within the food chain [4]. Literature reported that plants use up to 50% only of the applied fertilizer, 2 - 20% volatilized, 15 – 25% reacts with organic compounds in soil and 2 – 10% leach into groundwater and surface [5]. Large quantities of inorganic fertilizers application

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also led to severe water pollution where the excessive applied nitrogen, N may be potentially leached from the root and ends up in lakes, ponds, rivers and other water sources that results in an adverse effect [6]. Therefore, it is important for the farmers/growers to follow the guidelines and recommended dose of fertilizers usage issued by the Malaysian Department of Agriculture since the outcome from an observation conducted showed that many of the farmers applied more than the recommended dose. Hence, it is important to optimize the application of fertilizers in order to sustain agricultural production, so that the impact towards the environment could be minimized [7].

Therefore, studies on the optimization condition of AB mix fertilizers on *C. frutescens* plants have been carried out using the wick fertigation technique (Nutri-pot) via response surface methodology, (RSM) from week 1 to week 7 by utilizing a soft cloth thread that is used to absorb water and nutrients. RSM has been reported as an effective tool used by researchers to determine the optimization of a process. It is a collection of statistical and mathematical systems that are used in developing, improving and optimizing different systems even when the independent variables have a combined effect on the desired response [8]. Chemicals used in this study are the inorganic fertilizers, known as AB mix fertilizers which are commonly used in urban farming [9,10]. It consists of 2 functional solutions: solutions A and B where both contain macro- and micro-components [11] that are immediately available in providing nutrients to crops.

The present studies reported results of the optimum condition level of the height of *C. frutescens* plants from week 1 to week 7 using the Nutri-pot system. As far as we concern, no reports have been published on the optimization studies towards *C. frutescens* plant as most of the studies were reported on leafy vegetables such as pakchoy, lettuce and spinach. In this work, only two (2) factors are varied i.e. the concentration level (EC) of AB mix fertilizers and the wick length of the Nutri-pot. Comparison data between the simulated and the actual response of the plant's growth is also determined.

2. Methodology

2.1 Experimental Design

Materials used for plant preparation were AB mix fertilizers, coco-peat as medium, *C. frutescens* seedlings, aluminium foil, 5-liter bottles and water. The fertilizers and seedlings are complimentary from E-Office Agrotech Plantation Sdn. Bhd. The Nutri-pots were first constructed as in Figure 1 by cutting the 5 liter bottles into half. The upper sides of the containers were filled with coco-peat whilst, the bottom parts were the AB mix fertilizers, prepared at different EC concentrations of 1.8, 2.2 and 2.6 mS/cm. The ratio for parts A and B are similar for all plant samples with 1:1 each. The coco-peat was prepared by flushing it with water prior to the seedlings' transplant into the containers, provided with different lengths of wick: 17, 19 and 21 cm that will absorb the AB mix nutrients. Next, the fertilizers were poured into the bottom part of the containers and covered with aluminium foil to hinder algae growth, as shown in Figure 2. This methodology has been drawn from previous studies where the selected parameters were shown to be sufficient in achieving the objective of the studies [1]. The amount of AB mix fertilizers applied were similar for all samples at 500 ml. The grow and care of *C. frutescens* plants have been carried out in controlled environment at level 4, Bangunan Jauhari, away from extreme weather and pest-free area.

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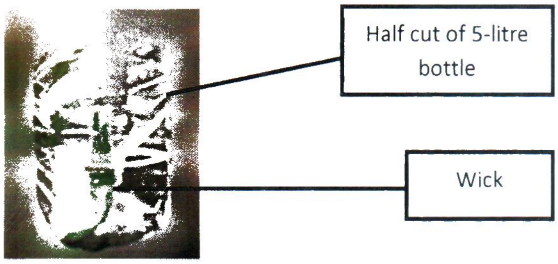

Fig. 1 Nutri-pot System

Fig. 2 Nutri-pot with *C. frutescens* plant covered with aluminium

In the first four weeks, all seedlings were introduced and fed with lower EC concentrations at 1.0, 1.2 and 1.6 mS/cm to avoid stunted growth, plant diseases and excessive feeding to the seedlings as shown in Table 1. At week 3 and 4, the EC concentrations were remained constant due to the burn effect observed in most of the tips of *C. frutescens* seedlings. Feeding the seedlings with lower amounts of fertilizer is necessary due to their sensitivity towards excessive nutrients, which can harm them. Lower concentrations of fertilizers reduce the risk of tip burn, stunted growth, nutrient imbalances, and leaf scorching [12]. Mixture of organic pesticides containing chili and garlic blend was applied to the plants one time per week to treat and protect the plants from whiteflies and pests.

Table 1 The growth of *C. frutescens* from week 1 - 4 for 19 cm wick length at different EC application

	Images of *C. frutescens*			
	Week 1 **EC level: 1.0 mS/cm**	**Week 2** **EC level: 1.2 mS/cm**	**Week 3** **EC level: 1.6 mS/cm**	**Week 4** **EC level: 1.6 mS/cm**
Sample #2 **Wick length: 17 cm**	 Height: 19.8 cm	 Height: 21.6 cm	 Height: 22.1 cm	 Height: 22.8 cm
Sample #10 **Wick length: 19 cm**	 Height: 27.1 cm	 Height: 28.7 cm	 Height: 31.9 cm	 Height: 33.6 cm
Sample #12 **Wick length: 21 cm**	 Height: 24.4 cm	 Height: 25.0 cm	 Height: 25.5 cm	 Height: 26.4 cm

2.2 Mathematical Model and Analysis

Statistical methodologies in different fields of research have been used for optimization. A conventional method of RSM is useful in modeling and analyzing situations in which, several variables influence a response of interest and the objective is to optimize the responses. Hence, the goal of RSM is to explore a proper approximating relationship between the input variables and the output responses as well as, to find the optimum operating conditions for a system under investigation or within which operating requirements are satisfied [13]. The response surface can be expressed in Eq. 1 as follows:

$$y = f(X_1, X_2, \dots, X_k) \quad (1)$$

where y is the response and X_k is the independent variable (factor). A central composite design (CCD) with two independent variables (X_1 for EC values of AB mix fertilizers and X_2 for wick length) at three levels were performed by applying the Design-Expert software package (Version 13.0.5, Stat-Ease Corp., US), whilst the crop height, Y_1: cm were studied as the response variable for the modelling. The height of the plants were measured using rope and measuring tape for data analysis. Each independent variable of three levels was coded as low (-1), centre point (0) and high (+1) with the two independent factors i.e. concentration level of AB mix fertilizers, X_1: 1.8, 2.2, and 2.6 mS/cm and wick length, X_2: 17, 19, and 21 cm were considered. To determine the number of CCD runs, the standard 2^k factorial with its origin at the centre is included. $2k$ points are fixed axially at a distance from the centre to generate the quadratic terms and replicate runs at the centre (r), where k is the number of variables. Hence, the total number of runs required for the two independent variables can be calculated using Eq. 2:

$$2^k + 2k + r = 22 + (2 \times 2) + 5 = 13 \quad (2)$$

Therefore, total of 13 experimental runs were required for the two independent variables. Tables 2 and 3 provide thorough explanation of the variable configuration along with their coded and actual levels of the EC values and wick length. The configuration of which model to use, the predictive models and their optimization were built by the chosen variables in Eq. 3 which expresses the equation of the second-order quadratic model:

$$Y = \beta_0 (\beta_i \times X_1) + (\beta_j \times X_2) + (\beta_{ij} \times X_1 \times X_2) + (\beta_i \times \beta_{12}) + (\beta_i \times X_{22}) \quad (3)$$

where, Y is the response in terms of predicted plant height, β_0, β_i, β_j, and β_{ij} are the interactive regression coefficients and X_1 and X_2 are the EC level of AB mix fertilizers and wick length, respectively. Adequacy and statistical significance of the variables in the model used were examined using ANOVA software by using the lack-of-fit test of different models. The coefficient of determination, R^2 was determined in order to assess the goodness and adequacy of the fitted model. Design Expert 13.0.5 software was used for regression analysis and graphical optimization.

Table 2 Experimental factors and level of CCD for different concentration levels (EC) of AB mix fertilizers and the wick length

Factors	Symbols	Coded levels		
		Low (-1)	Centre point (0)	High (+1)
EC value (mS/cm)	X_1	1.8	2.2	2.6
Wick length (cm)	X_2	17	19	21

Table 3 The coded and actual levels of independent variables

No. of runs	Coded levels		Actual levels	
	X_1	X_2	X_1	X_2
	EC level (mS/cm)	Wick length (cm)	EC level (mS/cm)	Wick length (cm)
1	0	0	2.2	19
2	0	-1	2.2	17
3	0	+1	2.2	21
4	+1	-1	2.6	17
5	-1	0	1.8	19
6	0	0	2.2	19
7	+1	0	2.6	19
8	+1	+1	2.6	21
9	0	0	2.2	19
10	0	0	2.2	19
11	-1	-1	1.8	17
12	-1	+1	1.8	21
13	0	0	2.2	19

3. Results

3.1 Effect of Different EC Values on C. frutescens' Height

Results of *C. frutescens*' height after seven weeks according to the 13 runs at different EC levels of AB mix fertilizers and wick length using CCD are shown in Table 4. It can be observed that the heights were significantly varied with the factors. The actual plants' heights ranged from a minimum of 25.5 cm to the maximum, 41.0 cm. The lowest height of 25.5 cm was observed in the maximum (+1) level of EC and wick length values of 2.6 (mS/cm) and 26 cm, respectively whereas, the maximum height, 41.0 cm was observed as the EC level and wick length were at medium (0) level of 2.2 mS/cm and 19 cm, respectively. All central points, (0, 0) had plant heights ranging from 39.0 cm to 39.8 cm whilst the axial (0, -1), (0, +1), (-1, 0), (+1, 0) and factorial (+1, +1), (+1, -1), (-1, -1), (-1, +1) points recorded lesser values. This revealed that the maximum height of *C. frutescens* can be achieved when both factors are at the appropriate amount of values.

Plant's height is one of the crucial parameters that are often used as a measure to indicate the crop's development therefore, any independent variables need to be at the correct amount. The EC level of AB mix fertilizers with medium (0) treatment helps the plant during the nutrient uptake process whilst, excessive or higher levels of EC value often contribute to brownish curled and shedding leaves [14]. Overuse of inorganic fertilizers has caused serious environmental pollution including deterioration of soils' fertility and their natural properties, and groundwater pollution which, therefore, contributes to global warming issues [15]. Similarly, the wick length of the Nutri-pot system used in this experiment is vital since it absorbs water and nutrients from the solution and supplies them to plants [16].

Table 4 Predicted and experiment response values of *C. frutescens*' height

No. of runs	X_1	X_2	Response Y_1	
			Δ Plant Height (cm)	
	EC level (mS/cm)	Wick length (cm)	Actual value	Predicted value
1	0	0	39.80	39.40
2	0	-1	41.00	41.07
3	0	+1	34.30	34.61
4	+1	-1	39.90	39.64
5	-1	0	36.70	36.44
6	0	0	39.20	39.40
7	+1	0	33.30	33.94
8	+1	+1	25.50	25.12
9	0	0	40.20	39.40
10	0	0	39.00	39.40
11	-1	-1	33.90	34.09
12	-1	+1	35.60	35.67
13	0	0	39.20	39.40

This approach may be an effective substitute with lesser environmental damage to enhance the quality of the plants, enabling production with reduced labour and electricity or in regions with high air temperatures [17]. It is crucial to emphasize that this analysis was conducted in a controlled environment with steps taken to eliminate potential pest attacks and the influence of extreme weather conditions.

3.2 Model Fitting

To examine the type of model used and the significance of each model, tests using different models namely linear, 2FI, quadratic and cubic have been carried out to find the relationship within the variables. As shown in Table 5, the coefficient values of linear and 2FI models were not significant as both show negative interactions from their values, whilst, for quadratic and cubic models, the values are significant. Based on the results, it can be observed that the quadratic model will be used as it gives positive values with the significant term in comparison with other models. After denoting that the best model is quadratic model, independent variables were fitted in the specified model and the effect of each variable was assessed. Therefore, several parameters were used as indicators to evaluate the adequacy of the fitted model, and the indicators were the coefficient of determination, (R^2), the adjusted determination coefficient, (adjusted R^2) and the predicted determination coefficient, (predicted R^2). Results shown in Table 6 indicates that the *F*-value for this model is 163.54. *F*-value is a measure of separation between various distributions during the experiment i.e. the variance of the data that can be analyzed and therefore, it is used to identify whether the test is statistically significant or vice versa. Therefore, the F-value of 163.54 implies that there is only a 0.01% chance that a model F-value this large could occur due to noise [18].

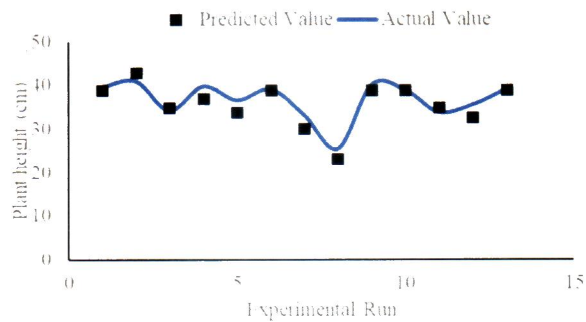

Fig. 3 Run-wise predicted and actual values of *C. frutescens* growth

Table 5 Analysis of variance for different models

Models	Sequential *P*-value	Lack of Fit *P*-value	R^2 Adjusted	R^2 Predicted
Linear	0.1376	0.0003	0.1930	-0.5937
2FI	0.0266	0.0006	0.4957	-0.7081
Quadratic	<0.0001	0.4345	0.9854	0.9568 (suggested)
Cubic	0.3447	0.4105	0.9867	0.8812 (aliased)

The F-value of independent variables i.e. *A*, *B*, *AB*, A^2 and B^2 are 35.12, 234.96, 242.74, 183.55 and 25.24, respectively which implied that the values and interaction effect on dependent variable are significantly high. The F-value is indirectly proportional to the *P*-value where *P*-value is a method used to quantify the probability that one can navigate the difference which could be the result of randomized tests. Generally, the statistical significance of the observed difference increased as the *P*-value decreased. Value less than 0.0500 indicates that the model term is significant. In this case, the independent variables of *A*, *B*, *AB*, A^2 and B^2 have a significant model term which is 0.0006, < 0.0001, < 0.0001, < 0.0001 and 0.0015, respectively. Equation 4 is the final equation in terms of actual factors fitting the model prediction.

$$\Delta \text{ Height} = -401.703 + 208.310 A + 24.2917 B + (-5.03125 AB) + (-26.3254 A2) + (-0.390517 B2) \quad \text{(Eq. 4)}$$

The *P*-value induced cvalue to be relatively high, 0.9915 as shown in Table 6. The value, 0.9915 denoted that approximately 99.15% of the variability observed in the target variable was explained by the regression model. If the R^2 value is low, the model does not explain much about the data variation. R^2 is a statistical measurement that shows how well the model fits the data or how much variation was explained by the model [18]. It also explains the extent to which the variance of one variable will be presented to the second variable even though the correlation explains the strength of the relationship between independent and dependent variables. The predicted R^2 value, 0.9568 was in reasonable agreement with the adjusted R^2 of 0.9854 where the difference is less than 0.2.

As for adequate precision value, it measures the signal-to-noise ratio where a ratio greater than 4 is desirable. As denoted in Table 6, the value of 45.450 indicates an acceptable signal. The lack of fit value, 1.14 denoted that the value is significant however, the non-significant lack of fit, 43.45 % denoted that the value of this large could occur due to the noise. Residual standard error, (RSE) data displayed 0.14 %, which indicates a smaller error percentage between the actual and predicted values [19].

The experimental data of the plant's height were compared with the predicted values using the second-order quadratic model. It can be observed in Figure 3 that the experimental values data fit well with the predicted data and there is a good correlation between the experiment and predicted values. The fitting is relatively good which demonstrates that the CCD model with an experimental design can be effectively applied for optimization [20].

Table 6 Analysis of variance for the quadratic model

Sources	**Sum of Squares**	***DF***	**Mean Square**	***F*-value**	***P*-value**
Model	218.30	5	43.66	163.54	<0.0001 significant
A (EC value)	9.38	1	9.38	35.12	0.0006 significant
B (wick length)	62.73	1	63.73	234.96	<0.0001 significant
AB	64.80	1	64.80	242.74	<0.0001 significant
A^2	49.00	1	49.00	183.55	<0.0001 significant
B^2	6.74	1	6.74	25.24	0.0015 significant
Residual	1.87	7	0.2670		
Lack of Fit	0.8608	3	0.2869	1.14	0.4345 not significant
Pure Error	1.01	4	0.2520		
Cor Total	220.17	12			
RSE (%)	0.14		Std. Deviation (√RSE)		0.5167
R^2	0.9915		Predicted R^2		0.9568
Adjusted R^2	0.9854		Adequate Precision		45.4496

Figure 4 shows the response surface and their corresponding contours of the combined effect of the EC value of AB mix fertilizers and wick length on the height of the *C. frutescens* plant. It is

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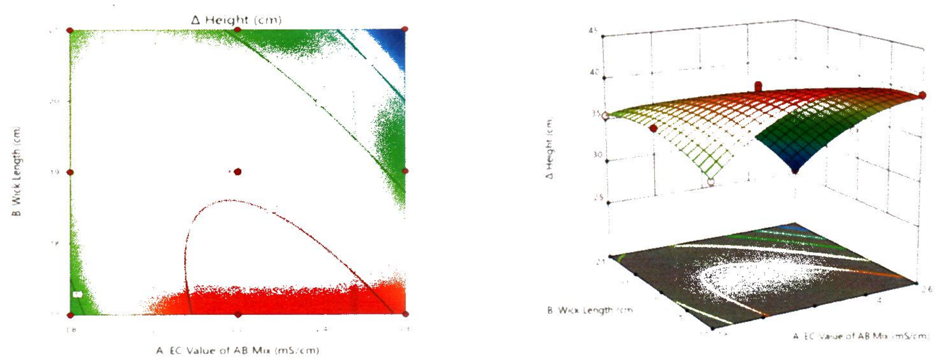
observable that sufficient amount of EC values and wick length can contribute to the height of the *C. frutescens* plant. Based on the contour plot graph in Figure 4 (a), darker orange-red regions possess maximum *C. frutescens* height (cm) whereas for yellow, green and blue regions indicate lower value of factors and response, consecutively. It can be observed that the maximum point was achieved when the EC value and wick length were 2.2 mS/cm and 19 cm, respectively at the height of *C. frutescens* was 40.2 cm.



(a) (b)

Fig. 4 (a) Response Contour Plot and (b) 3D Surface Plot for interactive effects of EC level of AB mix fertilizers and wick length

Based on the suggested quadratic model, Design-Expert software was used for optimizing the condition of AB mix usage on *C. frustescens* plant. The optimization studies were performed to evaluate the optimal experimental and predicted parameters. Table 7 compares the variables of the experiment along with the predicted and model-optimized values. The experimental data recorded that the highest plant was at 40.20 cm, compared to the predicted value, 39.40 cm as the value of operating parameters were at 2.2. mS/cm and 19 cm for EC level and wick length, respectively. Under the same conditions, the maximum height of *C. frustescens* plant was predicted to be 39.40 cm. The small difference between the predicted and actual values of the response has successfully verified the optimum point determined by RSM. The optimum values were found to be at the wick length and EC level of 18.15 cm, and 2.54 mS/cm, respectively where under the optimal conditions, the height of *C. frustescens* plant was expected to be 38.73 cm, which fitted well to the corresponding experimental value. According to the experimental data, Table 4, Run 4 displayed the nearest value to the optimum condition under the operating parameters of 2.6 mS/cm and 17 cm for EC level and wick length, respectively. The recorded data of the growth and development of *C. frutescens* plant in Nutri-pot at different EC application from Week 5 to Week 7 is shown in Appendix for reference.

Table 7 Comparison between the experiment, predicted and optimized values of RSM for *C. frutescens*

Variables	Experiment value	Predicted value	Optimized value using RSM
EC level (mS/cm)	2.2	2.2	2.54
Wick length (cm)	19	19	18.15
Δ Plant height	40.20	39.40	38.73
Plant height Deviation (%)	-	2.03	3.80

4. Conclusions

Optimizing condition of AB mix nutrient on *C. frustescens* plants in Nutri-pot from week 1 to week 7 has been carried out using response surface methodology, RSM. The statistical analysis based on the central composite design using quadratic model showed that at 2.54 mS/cm and 18.15 cm for EC level and wick length, respectively was the best condition to achieve the optimum *C. frutescens'* height. Compared to the experimental output, *C. frutescens'* height was recorded at 40.2 cm at the EC level and wick length of 2.2 mS/cm and 19 cm, respectively. This indicates the values of the parameters used are in the range of the optimization target. Overall, the operational parameters were successfully optimized using the proposed model. The application of prediction models in the industrial and agriculture sectors can assist in the advancements and emergence of technologies that contributed to significant improvement in the industries [21].

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