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Research article

Solutions for effective prevention of after-cooking discoloration in deep-fried eggplant (*Solanum melongena* L.)

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Abstract

Importance of the work: Deep-fried eggplant often undergoes the negative impact of after-cooking discoloration (ACD). The pretreatment method and deep-frying parameters are critical factors that can minimize ACD in deep-fried eggplant.

Objectives: To investigate the effects of different calcium chloride soaking and deep-frying conditions on the after-cooking discoloration of deep-fried eggplant.

Materials & Methods: A series of preliminary experiments was conducted to investigate the effect of calcium chloride concentration, soaking time, frying temperature and frying time on the ACD of deep-fried eggplant and identify the important factors. Frying temperature and frying time were chosen for further optimization study using response surface method (RSM).

Results: All the mentioned factors had positive effects on mitigating the ACD. Overall, the results demonstrated that pretreatment using calcium chloride helped to minimize the ACD of deep-fried eggplant, with the appropriate concentration and soaking time being 2% and 5 min, respectively. The results indicated that frying temperature and frying time were the most important parameters affecting the response. The RSM optimization results showed that the minimum color difference could be obtained at the optimum frying conditions of temperature = 161°C and time = 209 seconds and their factor contributions were 22 and 78%, respectively. The achieved color difference was about 3.48, which was close to 2.3, the "just noticeable color difference".

Main finding: The results provide a strategy for the development of industrial-scale production of deep-fried eggplant.

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Introduction

Solanum melongena L., commonly known as eggplant in the USA and Australia or aubergine in UK and France and brinjal in South Asia and South Africa, is one of the most commercially important vegetable crops and has been cultivated in various countries around the world since ancient time (Daunay and Janick, 2007). The production and gross value of eggplant reached 54 million t (2018) and USD 2.35 billion (2016) worldwide (Food and Agriculture Organization of the United Nations, 2020). There are various cultivars of eggplant, which are differentiated based on size, shape and weight (Sękara et al., 2007). Many of them contain large amounts of nutrients along with phytochemicals that are favorable to human health (Hanson et al., 2006). Eggplant needs to be cooked by heating before human consumption, with deep-frying being one of the most common processing methods that can be applied in both home preparation and industrial production.

One major difficulty in the eggplant deep-frying process is controlling the after-cooking discoloration (ACD) or browning caused by the activity of polyphenol oxidase (PPO, EC 1.14.18.1) (Banjongsinsiri et al., 2004). This enzyme catalyzes the oxidation of eggplant's phenolic compounds to *0*-quinones, which then build up and develop dark pigments, such as melanin (Uscanga-Sosa et al., 2020). The impact of heat in the frying process has been shown to have an inhibitory effect on PPO (Ayustaningwarno et al., 2018). However, this method is often impractical due to the requirement of a costly vacuum frying system to reduce the frying temperature and minimize the contact between frying products and oxygen in the air. Various product pretreatments prior to processing to prevent ACD have been investigated, with a considerable amount of study on the usage of calcium chloride as a pretreatment solution. (Guzek et al., 2012) reported the combination of calcium chloride soaking (1% mass per volume, m/v concentration) and low temperature blanching helped to significantly reduce the browning of broccoli. Likewise, studies have been carried out on the application of calcium chloride to prevent browning of fresh-cut products, such as with peach (Techakanon and Barrett, 2017) and pineapple (Gu et al., 2020).

Nuevo et al. (2020) reported the application of calcium chloride in combination with ascorbic acid to prevent browning in fresh-cut eggplant. However, it has not yet been established whether calcium chloride can minimize the ACD of deep-fried eggplant. Therefore, the aim of the current study was to investigate the effects of different calcium chloride soaking and deep-frying conditions on the after-cooking discoloration of deep-fried eggplant. Furthermore, response surface methodology was applied to determine the optimum conditions of frying temperature and frying time, as well as the contribution by each variable. Finally, the optimized process of deep-frying eggplant with minimum ACD was established for industrial scale application. The findings in this study should help to promote a simple solution to effectively prevent ACD in deep-fried eggplant.

Materials and Methods

Materials

Round, ripe eggplants (*Solanum melongena*, Ratna variety) were purchased from a local supermarket in Vietnam and stored at 4 °C until used. Calcium chloride ($CaCl_2$) anhydrous powder 98% purity was purchased from Merck, Germany. Water was obtained from a Milli-Q water purification system (Millipore; Bedford, MA, USA). All other reagents were of the highest grade commercially available.

Preparation of eggplant

Eggplants with similar size and weight were evaluated to remove defective items, with those remaining then being washed and the stem removed. Each round fruit was cut in half and each half was crosscut to create eight identical pieces with a pyramid shape and weighing 25 g on average. To prevent oxidation and browning, the fruits were cut shortly before pretreatment (Ioannou and Ghoul, 2013).

Pretreatment method of eggplant pieces

The pretreatment process was carried out as described by Techakanon and Barrett (2017) with some modifications. In brief, calcium chloride was dissolved in water to create pretreatment solutions with different concentrations. The freshly cut eggplant pieces were soaked in separate pretreatment solutions for different amounts of time with three replicates. After soaking, the pretreated pieces were dried in room temperature for 5 min prior to deep-frying. The eggplant pieces that had been soaked in water without calcium chloride were later used as negative control.

Deep-frying process of eggplant pieces

The treated eggplant pieces were deep-fried according to the process described by Khan et al. (2019) with some modifications. Briefly, a commercial Teflon-coated saucepan was filled with 2 L of canola oil and preheated on an induction stove (ICB-6619; Tara Co. JSC; Ho Chi Minh, Vietnam) for best heat stability and heat distribution (Imaz et al., 2014). Consistently, the oil temperature was monitored using a handheld data logging thermometer and kept stabilized at several fixed temperatures for at least 5 min before the eggplant was put in. The treated eggplant pieces were fried immersed in oil. A stainless-steel strainer was used to remove the fried pieces from the oil at predetermined times and the samples were allowed to drip for a few seconds before being cooled at ambient temperature on kitchen paper tissue. After cooling, the fried pieces were preserved at 0 °C for 48 h.

Color characteristics of deep-fried eggplant

The color change of the deep-fried eggplant pieces with different pretreatment and frying conditions before and after 48 h preservation was measured as described by Chitrakar et al. (2019). A handheld colorimeter (CR-400; Konica Minolta Sensing Americas Inc.; Ramsey, NJ, USA) was used to measure the CIE color values of L^* (lightness), a^*(redness) and b^* (yellowness). The colorimeter was first calibrated using the standard plate. The measurements were made at three randomly selected spots and the average color value was presented (Su et al., 2016). The color difference (ΔE^*) was calculated from the differences in color values of sample before preservation (L^*_1, a^*_1,b^*_1) and after preservation (L^*_2, a^*_2,b^*_2) using Equation 1:

$$\Delta E^* = \sqrt{(L^*_2 - L^*_1)^2 + (a^*_2 - a^*_1)^2 + (b^*_2 - b^*_1)^2} \quad (1)$$

Preliminary experiments design

A series of single-factor, completely randomized experiments was conducted prior to the optimization experiment with the aim of identifying important factors and narrowing the condition ranges. The independent variables studied were: calcium chloride concentration (0%, 1%, 2%, 3% or 4% m/v), soaking time (5 min, 10 min, 15 min or 20 min), frying temperature (140 °C, 160 °C, 180 °C, 200 °C) and frying time (30 s, 60 s, 120 s or 180 s). Color difference (ΔE^*) was the response variable. After each experiment, the condition level resulting in the minimal color difference was chosen as the control variable for the next experiments.

Optimization experiment design

The MODDE Design of Experiments software (version 12.1.0; MKS Data Analytics Solutions, Umeå, Sweden) was used to optimize the two most important factors affecting the ACD of deep-fried eggplant using the central composite design (CCD) method. The two selected independent variables were A: the frying temperature (X_1, 150–170 °C) and B: the frying time (X_2, 150–210 s) in accordance with the preliminary experiments. These variables were studied at three different levels coded as -1 (minimum), 0 (central) and +1 (maximum); the color difference (ΔE^*,Y) was chosen as the response variable of the system. In the MODDE optimizer objective, the color difference target was set to range from 0 to 10.0 on the basis of preliminary experiments to narrow down the optimized condition range.

The CCD model was applied to fit a second-order model requiring the minimum number of runs for modelling (Kumar et al., 2009). The CCD model consisted of 2^k factorial points with 2k axial points (where k is the number of parameters) and N_o central points (Dil et al., 2016). The number of runs (N) was calculated using Equation 2)

$$N = 2^k + 2k + N_0 \quad (2)$$

where k is the number of parameters and N_0 is the number of central points.

The actual (X) and coded (x) variable values are presented in Table 1. Three replicated central points were used for data reproducibility and experimental error determination (Toudeshki et al., 2019). Therefore, the CCD was designed based on carrying out four factorial points, four axial points and four replicates at the center (n = 12). The experimental run order was randomized to prevent systematic errors.

The value of the color difference (Y) was related to the coded variables (xi, i = 1 and 2) by a second-degree polynomial using Equation 3:

$$Y = a_0 + a_1x_1 + a_2x_2 + a_{12}x_1x_2 + a_{11}x_1^2 + a_{22}x_2^2 \quad (3)$$

where Y is the dependent response and the coefficients of the polynomial are represented by a_0 (constant term), a_1 and a_2 (linear effects), a_{11} and a_{22} (quadratic effects) and a_{12} (interaction effects).

Table 1 Observed and predicted results of central composite design to estimate effects of frying temperature and frying time on after-cooking discoloration in deep-fried eggplant

Run no.	Independent variable		Dependent variable	
	Frying temperature (°C)	Frying time (s)	Color difference (ΔE^*, Y)	
	X_1 (x_2)	X_2 (x_2)	Observed	Predicted
1	150 (-1)	150 (-1)	9.04	8.91
2	160 (0)	180 (0)	4.46	4.48
3	170 (+1)	150 (-1)	7.68	7.66
4	160 (0)	180 (0)	4.68	4.48
5	160 (0)	180 (0)	4.56	4.48
6	150 (-1)	210 (+1)	7.80	7.67
7	160 (0)	210 (+1)	3.39	3.54
8	160 (0)	180 (0)	4.50	4.48
9	150 (-1)	180 (0)	8.26	8.52
10	170 (+1)	210 (+1)	6.04	6.02
11	170 (+1)	180 (0)	7.03	7.07
12	160 (0)	150 (-1)	4.83	4.98

X = actual level of variables; x = coded level of variables

The regression analysis and three-dimensional (3D) response surface contour plots of the independent and dependent variables were used to estimate the optimum frying conditions. For further validation, the probability of failure was calculated of the optimized model with different independent variable precisions where the variation of frying temperature was set at ±1 °C, ±5 °C, ±10 °C, ±15 °C and ±20 °C, when the frying time variation was set at ±1 s. In contrast, the frying time variation was set at ±1 s, ±5 s, ±10 s, ±15 s and ±20 s while the frying temperature variation was set at ±1 °C.

Statistical analysis

All the experiments and analyses were done in triplicate, except for the optimization experiment. The data analysis was conducted using the Excel software (Microsoft Inc; Redmond, WA, USA) and the MODDE Design of Experiments software (version 12.1.0; MKS Data Analytics Solutions, Umeå, Sweden). All data were presented as mean values ± SD except for the optimization experiment. In the preliminary experiments, one-way analysis of variance (ANOVA) and Tukey's honestly significant difference test HSD were used to analyze the mean values at $p < 0.01$ (Wilkinson, 1989) to detect significant differences among levels of treatment. In the RSM-CCD experiment, ANOVA tables were developed to determine the effect and the regression coefficients of the individual linear, quadratic and interaction terms.

Results and Discussion

Preliminary screening studies were carried out on pretreatment conditions and frying conditions to identify the main factors affecting the ACD of deep-fried eggplant and to determine suitable value ranges.

Effects of pretreatment conditions

First, the effect was investigated of calcium chloride concentration on color difference before and after preservation. The cut eggplant pieces were soaked in different concentrations of pretreatment (calcium chloride) solution with the controlled variables (soaking time, frying temperature and frying time) kept at 10 min, 150 °C and 60 s, respectively, as described in Asadnahal et al. (2021). Fig. 1a shows the changes in color of the deep-fried samples before and after preservation. The effect of calcium chloride pretreatment in preventing discoloration and browning of processed food products during preservation has been reported for peach (Techakanon and Barrett, 2017), lettuce (Hengphum et al., 2015) and carrot (Yu et al., 2018). In the current study, the calcium chloride pretreatment was applied to deep-fried eggplant.

When the color difference (ΔE^*) was evaluated upon calcium chloride concentration, a decrease (Tukey's test, $p < 0.01$) was noted at the concentration of 2% (Fig. 1A), resulting in 14.32 color difference units, which was 21.3% lower than the control (18.19). The reduction of ACD with calcium chloride pretreatment was due to the inhibition of the PPO enzymatic browning activity by Cl- ions (Garcia and Barrett, 2002). In addition, the cell wall structure was strengthened and stabilized as the reaction progressed between the Ca^{2+} ions and native pectin of the eggplant tissue (Langer et al., 2019), restricting water transfer and making it harder for water to escape the cells (Modesto et al., 2020) and thus reducing the discoloration due to dryness during the preservation process (Nunes and Emond, 2007).

However, the elevated presence of the Ca^{2+} ions on the sample surface might have obstructed the Maillard reaction in the fried products (Lindsay and Jang, 2005). As reported by Vhangani and Wyk (2021), the products of the Maillard reaction caused a strong inhibitory effect on PPO enzymatic browning activity. Accordingly, mitigation of the Maillard reaction might resultin greater discoloration and browning in fried eggplant, which was evidenced by the increased color difference at a higher concentration of calcium chloride pretreatment solution (Fig. 1A). Considering the lowest color difference result, the pretreatment solution of 2% calcium chloride was chosen for further study on the effect of soaking time.

Second, the effect of the soaking time on the color difference was inspected. The cut eggplant pieces were soaked in the pretreatment (calcium chloride) solution with different soaking times of 5 min; 10 min; 15 min and 20 min. The controlled variables (calcium chloride concentration, frying temperature and frying time) were kept at 2%, 150°C and 60 s, respectively. The color differences of the samples before and after preservation were affected by calcium chloride pretreatment but varied significantly with soaking time (Fig. 1B). The color difference reached its minimum level at 5 min and increased with longer soaking times (Tukey's test, $p < 0.01$). This agreed with the earlier explanations, in which, calcium chloride prevented the discoloration and browning of deep-fried eggplant. However, an excessive amount of calcium chloride could bring unfavorable impacts due to the inhibition of the Maillard reaction leading to the stronger activity of PPO as previously interpreted. Additionally, there was a slight reduction in the color difference at 20 min.This could potentially be explained as the strong browning activity of PPO that happened during the frying and cooling times, which was accredited to the mildly lower lightness value (L^*, data not shown) of this sample before preservation, caused the overall reduction in the color difference after preservation.

Calcium chloride has been used as an anti-browning agent in several fresh or processed fruit and vegetable products (Antunes et al., 2010) owning to its various biological activities, with the chloride (Cl^-) ion being proven to have inhibitory activity against PPO (Garcia and Barrett, 2002), which is the enzyme responsible for browning and discoloration (Teoh et al., 2016). In addition, the complexity interactions between the Ca^{2+} ions and the miscellaneous pectin profiles in several plants, varieties and organs play an important role

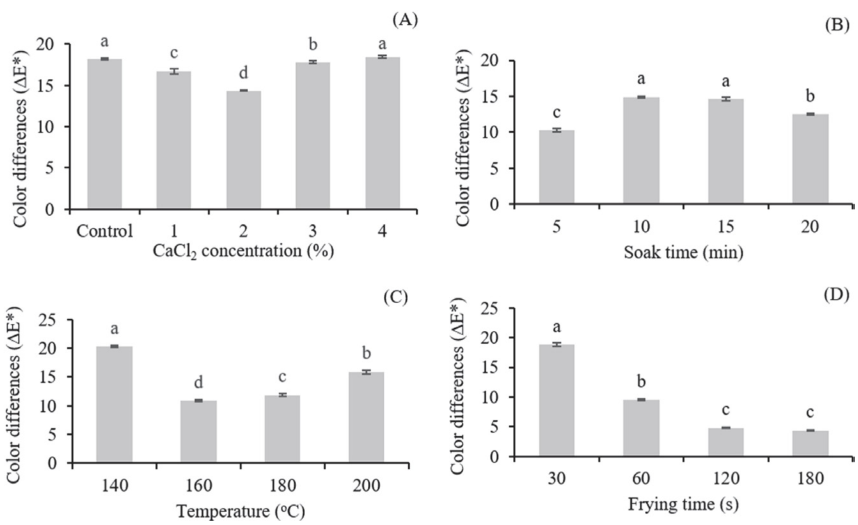

Fig. 1 Effect of: (A) $CaCl_2$ concentration; (B) $CaCl_2$ soaking time; (C) frying temperature; (D) frying time on color differences, where data are expressed as mean ± SD and different letters above bars indicate significant ($p < 0.01$) differences

in strengthening the cell structure and conserving the aqueous interior (Jain et al., 2019) and thus preventing discoloration due to dryness in preservation. Nevertheless, excessive increments in the calcium chloride concentration and soaking time led to a rise in color difference before and after preservation. Taking color difference minimization and industrial scalability into consideration, the authors decided to use calcium chloride concentration, a soaking time of 2% (m/v) and 5 min, respectively, for further studies.

Effects of frying conditions

First, the effect was investigated of frying temperature on color difference before and after preservation. The controlled variables (calcium chloride concentration, soaking time, frying time) were kept at 2%, 5 min and 60 s, respectively. Fig. 1C depicts the changes in color of deep-fried samples before and after preservation. There was a decline (Tukey's test, $p < 0.01$) in the color difference value from 20.31 to 10.31 when the temperature increased from 140 °C to 160 °C. This was in good agreement with another study (Todaro et al., 2011) that reported that the eggplant PPO was rapidly inhibited with an increase in the temperature, which might explain the reduction in color difference after preservation.

Nevertheless, further increment of the frying temperature to 180 °C and 200 °C resulted in slight intensification of the color difference (Tukey's test, $p < 0.01$). This suggested that the ACD of the deep-fried eggplant might be related to non-enzymatic browning, which was confirmed to be strongly associated with L-ascorbic acid (vitamin C) loss during the thermal process (Yang et al., 2021). Eggplant contains a moderate amount of L-ascorbic acid, with an average quantity of 89 mg/100g (Hanson et al., 2006). Lima et al. 2010 identified a significant degradation of L-ascorbic acid for a heat treatment higher than 160 °C. This contrast was a possible tentative explanation for the increase in the color difference in the samples fried at 180 °C and 200 °C. Considering the lowest color difference result, the frying temperature of 160 °C was chosen for further study on the effect of frying time.

Next, effect of frying time on color difference was examined, when the controlled variables (calcium chloride concentration, soaking time, frying temperature) were kept at 2%, 5 min and 160 °C, respectively. There was a significant decrease (Tukey's test, $p < 0.01$) in the color difference value from 18.85 to 4.34 when the frying time was extended from 30 s to 180 s (Fig. 1D), suggesting the substantial effect of frying time on the color difference of deep-fried eggplant before and after preservation. This was consistent with what has been found in previous studies, when increments of frying time were constantly associated with a decline in color change (Diamante et al., 2012; Eissa et al., 2013). In this case, a popular explanation of color difference reduction is that the longer frying time further promotes the Maillard reaction (Ciesarová et al., 2009), thus it forcefully inhibits the activity of PPO.

Optimization experiment

Subsequently, an optimization experiment was conducted to the previous screening studies. The important factors (frying temperature, frying time) were chosen for optimizing on account of their significant effect on reducing ACD in deep-fried eggplant.

First, using the MODDE software, the results of the 12 experimental runs were input and analyzed with the purpose of fitting a model using multiple linear regression. Table 1 shows the experimental design and results of 12 runs based on CCD. The regression coefficients of Equation 2 estimated by the software were: $a_0 = 4.48$, $a_1 = –0.73$, $a_0 = –0.72$ and $a_{11} = 3.3$. The coefficients of a_{22} and a_{12} were rejected because they had probability values (p-values) greater than 0.05, indicating not significant confidence levels. The coefficient of determination (R^2) and adjusted determination coefficient (adj. R^2) were calculated (0.995 and 0.991 respectively), thus indicating the high accuracy of the above model. In addition, there was good agreement between the calculated model for frying conditions and the experiment data. Final the color difference Equation 3 incorporates the frying conditions:

$$Y = 4.48 – 0.73x_1 – 0.72x_2 + 3.3x_1^2 \tag{3}$$

Second, the plot for observed (experimental) versus predicted results for color difference is shown in Fig. 2A. From the plot, the observed results were clearly relatively distributed close to a straight line, indicating good agreement of the observed and predicted results (good fit of data). This suggested that the fitted CCD model was reliable and could be effectively applied for controlling the ACD in eggplant the deep-frying process.

Third, the 3D surface contour plot (Fig. 2B) was used to investigate the relationship between the important independent factors and the response. As seen in the plot,

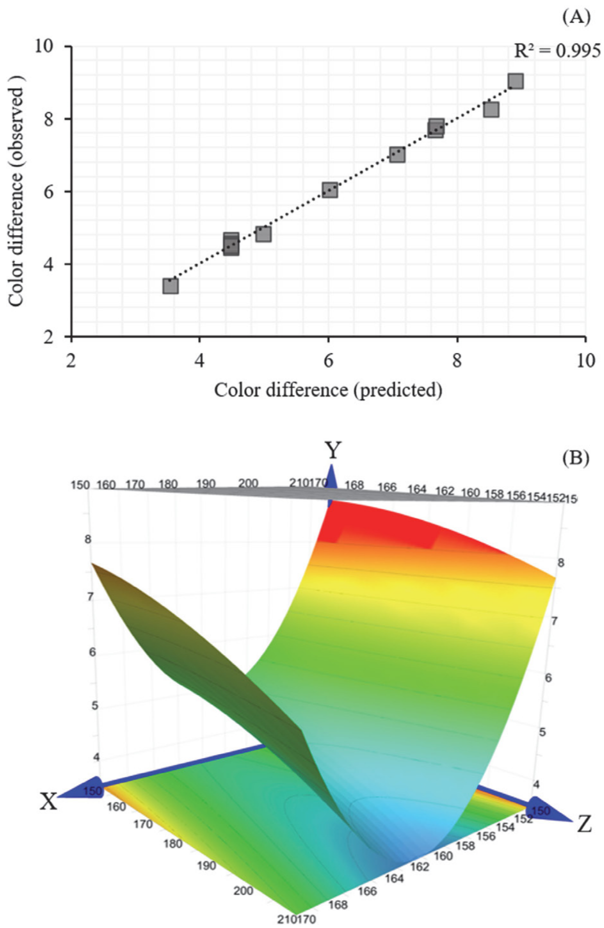

Fig. 2 Observed versus predicted values of color difference (A); 3D response surface plot of combined effects of frying temperature and frying time on color difference (B), where R^2 = coefficient of determination and X, Y and Z axes represent frying temperature (°C), color difference and frying time (s), respectively

when frying temperature increased toward 160 °C, the color difference decreased. A further increase in the temperature resulted in intensification of the color difference. In addition, there was a steady reduction in the color difference incorporation with frying time increment from 150 to 210 seconds. Such behavior was previously explained in the screening experiments, where enzymatic, non-enzymatic browning and Maillard reaction were taken into account. The main target of the optimization experiment was the achievement of a minimum color difference of deep-fried eggplant before and after preservation. The RSM optimization results showed that the minimum color difference could be obtained at the optimum frying conditions of temperature = 161 °C and time = 209 s, with their factor contributions being 22% and 78%, respectively. The achieved color difference was about 3.48, which was close to 2.3, the "just noticeable color difference" (Giorgianni et al., 2017), indicating that there was unnoticeable after-cooking discoloration of the deep-fried eggplant pieces after 48 h of preservation.

Finally, the model was further validated by estimating the probability of failure when applying the optimum conditions with different levels of precision of the independent factors. As seen in Fig. 3A, the probability of failure increased noticeably when the precision of the frying temperature decreased (from 0.01% up to 62% when the variation in frying temperature was raised from ±1 °C to ±20 °C). However, Fig. 3B shows that the probability of failure seemed to be stable with the lower precision of frying time, from 0.01% up to 9% when the variation of frying time increased from ±1 s to ±20 s. This suggested that although the frying temperature might have had a small contribution to the model (22%), it still needed to be well-handled in terms of precision in order to obtain a minimum color difference.

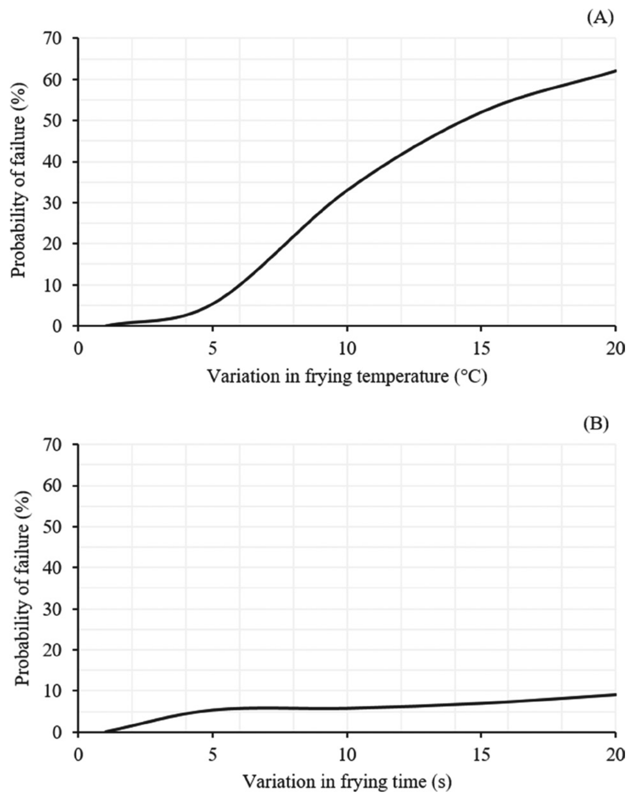

Fig. 3 Probability of model failure with variation in: (A) frying temperature; (B) frying time

Considering the results of the screening and optimization experiments along with practical application capability, an optimum process for deep-fried eggplant with the minimum ACD contained three-steps, as displayed in the flowchart in Fig. 4. Initially, the eggplant pieces are prepared by removing defective items, washed, the stem is removed and then the remaining sample is cut into eight identical pieces with an average weight of 25 g. Second, pretreatment is conducted by soaking the eggplant pieces in calcium chloride (2% concentration) for 5 min. Finally, the eggplant pieces are deep-fried at 160 °C for 210 s before further preservation.

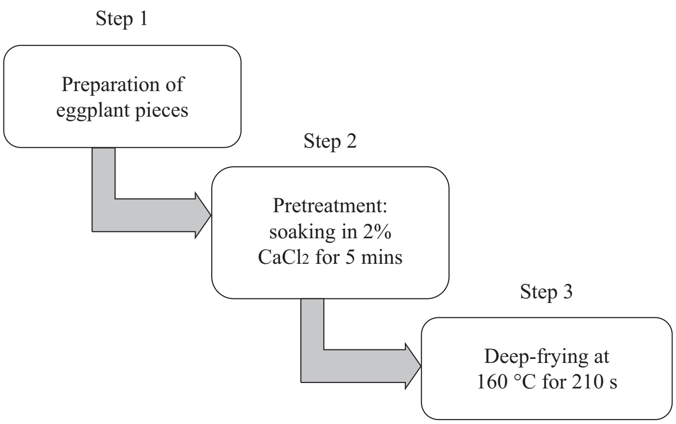

Fig. 4 Optimized process workflow for deep-frying eggplant

The effects of the different pretreatment and frying conditions on the after-cooking discoloration of deep-fried eggplant were successfully investigated and the frying conditions were further optimized based on the RSM-CCD method. Overall, the results demonstrated that pretreatment using calcium chloride helped to minimize the ACD in deep-fried eggplant, with the appropriate concentration and soaking time being 2% and 5 min, respectively. The frying conditions had a significant influence on reducing the ACD. The optimum conditions were at 161°C frying temperature and 209 s frying time. Finally, a process was proposed for deep-fried eggplant at a scalable level for industrial processing.

Conflict of Interest

The authors declare that there are no conflicts of interest.

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