International Food Research Journal 23(6): 2390-2398 (December 2016)
Journal homepage: http://www.ifrj.upm.edu.my

Optimization for anthocyanin and antioxidant contents and effects of acidulants on purple corn cake containing corn silk powder qualities

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Article history

Received: 18 July 2015
Received in revised form: 1 February 2016
Accepted: 14 February 2016

Keywords

Purple corn flour
Corn silk powder
Response surface methodology
Phytochemical compounds,
Antioxidant activities

Abstract

The objectives of this study were: 1) to investigate the optimal raw material ratio of purple corn cake containing corn silk powder (PCC) for maximal the total anthocyanin content (TAC), 2,2'-azinobis(3-ethylbenzothiazoline-6-sulfonic acid) radical scavenging ability (ABTS-RSA) and texture score using response surface methodology (RSM) and 2) to evaluate the effects of adding organic acids of PCC on TAC, total phenolic content (TPC), ABTS-RSA, ferrous chelating ability (FCA), color values and sensory evaluation. The optimal raw material ratio for maximal TAC (142.0 µg CE/g), ABTS-RSA (1,249.0 µg TE/g) and texture score (6.6) was the ratio of purple corn flour to rice flour of 71.1:28.9, corn silk content of 10.0 and water content of 27.1 (% total flour weight basis). The acidified PCC (APCC) with citric acid, fumaric acid, glucono-delta-lactone (GDL) and lactic acid at 0, 0.3, 0.75, 1.5, 3.0, 4.5 and 6.0% (% total flour weight basis) could reduce the degradations of TAC, TPC, ABTS-RSA and FCA. However, APCC with 3% fumaric acid showed the highest of TAC (282.7 µg CE/g), TPC (2,154.7 µg GE/g), ABTS-RSA (1,884.8 µg TE/g) and FCA (5,571.1 µg EDTA/g). The difference color (ΔE) and chroma values of APCC were significantly increased as increasing acid concentrations. Inversely, hue value was decreased as increasing acid concentrations. Additionally, the sensory scores in taste, texture and overall acceptability of APCC with 0.3% organic acid were higher than other acid preparations.

Introduction

Celiac disease or gluten-sensitive enteropathy (GSE) is identified by absorption disorders of small intestinal after taking the prolamins from wheat as well as other Triticum species (Turabi *et al*., 2008). Therefore, allergic gluten people are necessary to avoid the gluten diet. Maize (*Zea mays* L.) and rice *(Oryza sativa)* have very low gluten content (Turabi *et al*., 2008; De la Hera *et al.*, 2013) frequently used as a wheat substitute for gluten-free food products (Sivaramakrishnan *et al*., 2004). Nowadays, purple corn is widely cultivated in Thailand and continually developed and improved the genotype to increase the production and rich source of phytochemical compounds (Panrapee *et al.*, 2010). Anthocyanin content and the visible color of purple corn grains are associated with the aleurone layer of the endosperm (Kong *et al*., 2003). Moreover, purple corn silk is the one of a rich source of anthocyanin compounds (Sarepoua *et al*., 2013). The major anthocyanin compound is cyanidin-3-glucoside presented in the purple corn almost 75% of total anthocyanin compounds (Del Pozo-Insfran *et al*., 2006). Recently, anthocyanin compounds are more interest due to the health benefits and nutraceutical effects such as antioxidant properties, anticancer and anti-inflammatory activity (Tajkarimi and Ibrahim, 2011). Therefore, purple corn is the main source of low priced anthocyanin compounds compared with other phytochemical plants (Pascual-Teresa *et al.*, 2002).

Many factors in food processing such as oxygen, light, pH, sugar and temperature as well as storage condition significantly affect to the degradations of anthocyanin compounds (Sadilova *et al*., 2006). Additionally, many literatures reported that water activity (a_w) or water content is the major factor affecting on anthocyanin degradations. The antocyanin compounds are hydrolyzed by water with high temperature affected to a reduction of this compounds (Zazueta-Morales *et al.*, 2002). Sadilova *et al.* (2006) reported that anthocyanin compounds are significantly degraded as increasing a_w at 100°C. However, Del Pozo-Insfran *et al.* (2006) revealed that adding organic acids in products could significantly reduce the anthocyanin degradations.

To improve the bakery products for GSE people, the purple corn flour is interestingly used

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to substitute wheat flour as well as delivering the healthy compounds to the consumer (Li *et al.*, 2011). However, gluten in wheat is mainly source of protein structure which enhances the dough forming and viscoelastic properties of bakery products (Sivaramakrishnan *et al.*, 2004). Therefore, some food additives such as gums or hydrocolloids were added to the gluten-free food products to obtain the satisfying quality (Tajkarimi and Ibrahim, 2011). Guar gum is polysaccharide composes of long chain molecule and high molecular weight (Turabi *et al.*, 2008). This gum is usually added to the food products to improve the mouth feel and change the viscosity (Van *et al*., 2006).

There have been a few reports focusing on the optimal ratio of rice flour and purple corn flour to substitute wheat flour in cake products. Therefore, this research primary focused at the optimal raw material ratio of purple corn cake containing corn silk powder (PCC) for maximal the total anthocyanin content (TAC), 2,2'-azinobis(3-ethylbenzothiazoline-6-sulfonic acid) radical scavenging ability (ABTS-RSA) and texture score using response surface methodology (RSM). Finally, the secondary objective was to evaluate the effects of adding organic acids of PCC on TAC, total phenolic content (TPC), ABTS-RSA, ferrous chelating ability (FCA), color values and sensory evaluation.

Materials and Methods

Purple corn flour and corn silk powder preparation

Purple corn kernels (PCKs) and corn silk (CS) were cultivated and harvested at 60-70 days used in this study. These raw materials were purchased from Nakhon Phanom Province, Thailand. PCKs and CS were dried at 60°C for 24 hours (Hu *et al*., 2010) using a hot air oven (U30, Memmert, German). Dried PCKs were milled into whole-grain purple corn flour (PCF) by a disk mill (Disk Mill FFC-45, Qingdao King Lion Imp & Exp Co. Ltd, China) and then were sieved through 80 mesh screens (Kokkaew *et al*., 2015). Meanwhile, dried CS was grounded to powder using a grinder. PCF and CS powder were kept in a drying container before using to prepare the purple corn cake containing corn silk (PCC).

Purple corn cake containing corn silk powder process

The PCC was prepared according to the formula of Turabi *et al.* (2008) composed of 100% total flour containing a blend of PCF and rice flour (RF), 100% sugar, 25% shortening, 9% egg white powder, 3% salt, 5% baking powder, 1% guar gum and vary of CS and water content (WC) (% total flour weight basis). Firstly, all dry ingredients including mix flour, baking powder, salt, guar gum and CS were thoroughly mixed. Melted shortening was mixed with sugar and egg white powder at 85 rpm for 1 minute using a mixer (Kitchenaid 5K45SS, Kitchenaid Ltd., USA). After that, dry ingredient mix and water were added to the mixture and mixed with speed at 85 rpm for 2 minutes and then increased speed to 140 rpm for 1 minute and finally the mixer were decreased speed to 85 rpm for 2 minutes. The batter cake samples around 70-80 g were poured into the mould and baked at 185°C for 15 minutes using oven (Electrolux EOB64150X, Electrolux Thailand Co, Ltd., Thailand). After cooling down, PCC were vacuum-packed (Type SAE 10, Supervac Ltd., Germany) and immediately stored at -18°C (SF-C695, Sanyo Co. Ltd., Thailand).

The various effects in ratio of PCF to RF (PCF: RF), CS and WC (% total flour weight basis) were determined using RSM. The 23 central composite designs (CCD) were used to optimize the raw material ratio for maximal the TAC (Y_1), ABTS-RSA (Y_2) and texture score (Y_3) (Guo-qing *et al*., 2005). Symbols and coded variable levels for the variables (PCF:RF (x_1), CS (x_2) and WC (x_3)) were shown in Table 1. Meanwhile, the response surface were obtained using +|α|=1.68. In this study, CCD generated the 20 designed experiments (Table 2). The second-order model proposed for the response (Y) was:

$$Y = \beta_0+\beta_1x_1+\beta_2x_2+\beta_2x^3+\beta_{11}x_1^2+\beta_{22}x_2^2+\beta_{33}x_3^2+\beta_{123}x^1x^2x^3 \quad (1)$$

where Y is the response variable, x_1, x_2 and x_3 are the coded variables and β_0, β_1, β_2, β_3, β_{11}, β_{22}, β_{33} and β_{123} are the regression coefficients (Montgomery, 1991).

Acidulant addition in PCC

To improve the PCC qualities, acidulants including citric acid, fumaric acid, glucono-delta-lactone (GDL) and lactic acid were used to prepare the acidified PCC (APCC). Briefly, PCC containing 100% total flour (PCF:RF; 71.1:28.9), 100% sugar, 25% shortening, 9% egg white powder, 3% salt, 5% baking powder, 1% guar gum, 10% CS and 27.1% WC (% total flour weight basis). Acidulant was added with variation of concentration at 0, 0.3, 0.75, 1.5, 3.0, 4.5 and 6.0 (% total flour weight basis). After that, PCC was analyzed to evaluate the TAC, TPC, ABTS-RSA, FCA, color values and sensory evaluation.

pH measurements

The pH measurements on PCC were analyzed

Table 1. Independent variables and levels of CCD applied to PCC process

Independent variables		Levels				
Code	Real	-α	+1	0	+1	+α
x_1	PCF:RF (% total flour weight basis)	39.8:61.2	50:50	65:35	80:20	90.2:9.8
x_2	CS (% total flour weight basis)	3.3	5.0	7.5	10.0	11.7
x_3	WC (% total flour weight basis)	23.3	25.0	27.5	30.0	31.7

Table 2. Response values for Y_1, Y_2 and Y_3 with different combinations of x_1, x_2 and x_3 in CCD

Run	Code level			Response values		
	x_1 (%)	x_2 (%)	x_3 (%)	Y_1 (µg CE/g)	Y_2 (µg TE/g)	Y_3 (score)
1	65.0:35.0	7.5	27.5	123.5	980.1	7.2
2	65.0:35.0	3.3	27.5	100.0	922.8	7.5
3	80.0:20.0	10.0	30.0	142.4	1,253.8	5.2
4	65.0:35.0	7.5	27.5	131.6	1,085.4	7.5
5	39.8:60.2	7.5	27.5	80.5	905	7.2
6	65.0:35.0	11.7	27.5	137.6	1,258.6	6.6
7	50.0:50.0	5.0	30.0	98.1	756.4	7.8
8	50.0:50.0	5.0	25.0	86.8	940.9	6.9
9	90.2:9.8	7.5	27.5	152.4	1,255.5	5.3
10	65.0:35.0	7.5	23.3	101.1	954.1	6.5
11	50.0:50.0	10.0	25.0	106.0	898.1	6.6
12	50.0:50.0	10.0	30.0	122.6	895.2	6.9
13	80.0:20.0	10.0	25.0	136.6	1,280.7	5.9
14	65.0:35.0	7.5	31.7	110.9	824.1	6.2
15	80.0:20.0	5.0	30.0	128.6	875.5	6.7
16	65.0:35.0	7.5	27.5	133.9	1,164.3	7.2
17	65.0:35.0	7.5	27.5	121.6	1,139.1	7.1
18	80.0:20.0	5.0	25.0	106.2	952.4	5.3
19	65.0:35.0	7.5	27.5	130.5	1,049.9	7.0
20	65.0:35.0	7.5	27.5	129.2	1,050.5	7.2

*x_1: Purple corn flour: rice flour (PCF:RF), x_2: corn silk content (CS) x_3: water content (WC)
*Y_1: total phenolic content (TPC),Y_2: ABTS radical scavenging activity (ABTS-RSA) and Y_3: texture score

following the method of Li *et al.* (2011). Firstly, a PCC (10 g) was ground and dispersed in 100 ml of distilled water. The mixture was left at room temperature for 30 minutes and then measured pH using pH meter (pH211, Hanna Instrument Ltd., USA.).

Extraction of phytochemical compounds

The extraction of phytochemical compounds was performed according to the modified method of Abdel-Aal and Hucl (1999). Ground PCC (3 g) was weighed in a 50 ml centrifuge tube, and then 24 ml of methanol was added. The mixture was thoroughly shaken for 30 minutes using shaker (Nine, Wizard, Thailand) then centrifuged at 12,000 x g for 15 minutes (Centrifuge refrigerator, Avanti TM J25, Beckman Ltd., USA.). The supernatant or extract was evaporated to remove methanol using rotary evaporator (Buchi R-114, Buchi Corp., USA.) at 50°C. The extract was kept at -18°C and analyzed to determine the TAC, TPC, ABTS-RSA and FCA.

Determination of TAC

TAC was determined using the pH differential method according to the method of Wrolstad *et al.*

(1982). Meanwhile, cyanidin-3-glucoside equivalents used to quantify the TAC. The mixture solution was measured the absorbance with a UV-visible spectrophotometer (Lambda 25, Perkin Elmer, Inc., Germany) simultaneously at λ = 420 and λ = 700 nm in buffers at pH 1.0 and 4.5 and then A was calculated following: A = (A420 - A700) pH 1.0 - (A420 - A700) pH 4.5. Finally, TAC was calculated as follow: TAC = (A x MW x DF x 1000)/(ε x l) where, DF was dilution factor, ε was a molar absorption of 26,900 L/cm/mol, MW was molecular weight of 449.2 g/mol. TAC was expressed as µg CE/g dry sample.

Determination of TPC

TPC determination was performed with Folin-Ciocalteau reagent assay described by Inglett *et al.* (2011). A 200 µl diluted extract and 0.5 ml of Folin-Ciocalteau reagent (Folin-Ciocalteau reagent diluted with water, 1:1, v/v) were added into a test tube. The mixture was shaken and added with 0.8 ml of sodium bicarbonate (7% w/v). The mixture was left to stand at room temperature for 2 hours in the dark. The absorbance of the mixture was measured at 765 nm with UV-vis spectrophotometer against blank. TPC was compared with gallic acid standard curve and expressed as mg GAE/g dry sample.

Determination of ABTS-RSA

ABTS-RSA was analyzed according to the modified method of Re *et al.* (1999). To form $ABTS^{\bullet +}$, a 7 mM ABTS solution was oxidized with a haft volume of 2.45 mM $K_2S_4O_8$ for 12-16 hours in the dark. The $ABTS^{\bullet +}$ solution was diluted in 0.1 M phosphate buffer containing 0.818% NaCl and 0.0015% KCl, pH 7.4 before assay, giving an absorbance of 0.70+0.2 at 734 nm. A 200 µl diluted extract, 3 ml of $ABTS^{\bullet +}$ solution was added and then thoroughly mixed with vortex mixer. The mixture was incubated at room temperature for 6 minutes in the dark. Absorbance of mixture was measured at 734 nm UV-vis spectrophotometer. ABTS-RSA was determined by standard curve of Trolox (6-hydroxy-2.5.7.8 tetramethyl-chloroman-2-carboxylic acid) concentration versus ABTS-RSA (%) and expressed as mg TE/g dry sample.

Determination of FCA

FCA was performed by the modified method of Yin *et al.* (2005). A 200 µl diluted extract, was mixed with 3.7 ml methanol and 0.1 ml of 2 mM $FeCl_2.4H_2O$ and then uniformly mixed. The mixture obtained was mixed with 100 µl of 5 mM ferrozine and left at room temperature for 10 minutes. The absorbance was determined at 562 nm with UV-vis spectrophotometer. A standard curve of EDTA (ethylenediminetetraacetic acid) concentration and FCA (%) used to determine the FCA as µg EDTA/g dry sample.

Color measurements

Color characteristics of APCC were analyzed by Hunter CIE color (CIE, 2004) in Lightness *(L^*)*, redness *(a^*)* and yellowness *(b^*)* values using a chroma meter (Minolta CR-300 series, Minolta Co. Ltd., Japan). Chroma, hue, and E values were calculated using three equations: Chroma = $(a^{*2} + b^{*2})^{1/2}$; Hue = $\tan^{-1} (b^*/a^*)$; and E value = $(\Delta L^{*2} + a^{*2} + b^{*2})^{1/2}$.

Sensory evaluation of PCC

The sensory evaluation of PCC was performed to determine the color, taste, texture and overall acceptability scores using a nine-point hedonic scale (1 = extremely dislike, through to 9 = extremely like) with 50 untrained panelists.

Statistical analysis

CCD was conducted to maximize the TPC, ABTS-RSA and texture score of PCC using design expert software version 9 (Stat-Ease, Inc., Minneapolis, MN, USA.). Meanwhile, the effects of acidulants on PCC qualities were performed using the completed randomized design (CRD). Additionally, Duncan's multiple range test ($p<0.05$) used to determine the significant differences between mean values. Statistical analysis was carried out using SPSS statistic program (Version 17) for Windows (SPSS Inc., Chicago, IL, USA). All runs were performed in triplicate and data were averaged.

Results and Discussion

Central composite design (CCD)

The variation values of TAC (Y_1), ABTS-RSA (Y_2) and texture score (Y_3) in CCD were 80.5 to 152.4 µg CE/g, 756.4 to 1,280.7 µg TE/g and 5.2 to 7.8, respectively. The optimal of three variables to maximize the TAC (142.0 µg CE/g), ABTS-RSA (1,249.0 µg TE/g) and texture score (6.6) was PCF:RF of 71.1:28.9, CS of 10.0 and WC of 27.1 (% total flour weight basis).

The coefficient of determination (R^2) of three regression models (Y_1, Y_2 and Y_3) determined a degree of fit of model were 92.7, 94.0 and 92.8. Additionally, lack of fit for these model did not show significance (p=0.0981, 0.9089 and 0.7292). The results indicated that the adjusted model fit to the experimental data. The second-order models were

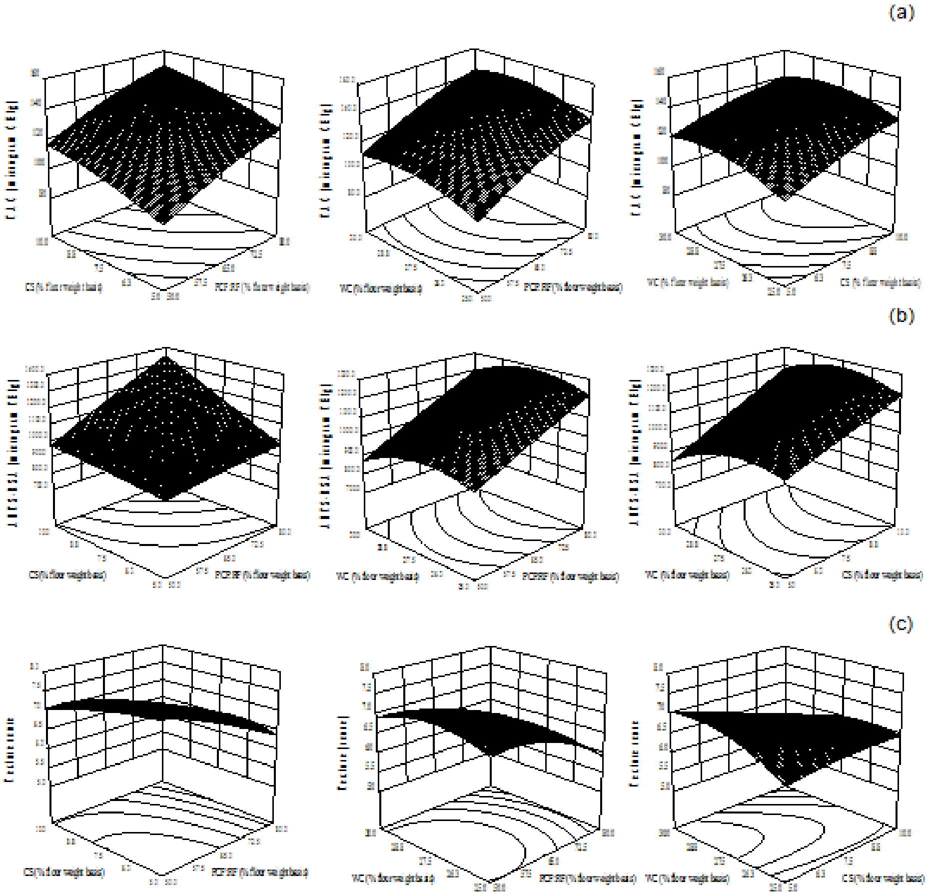

Figure 1. Three dimension response surface curves of interactive effects of purple corn flour: rice flour (PCF:RF), corn silk content (CS) and water content (WC) on (a) total anthocyanin content (TAC), (b) ABTS radical scavenging activity (ABTS-RSA) and (c) texture score

presented in Eq. (2), (3) and (4).

$$Y_1 = 128.3 + 16.2x_1 + 11.1x_2 + 5.3x_3 - 3.6x_1^2 - 2.7x_2^2 - 7.3x_3^2 + 0.1x_1x_2 + 0.04x_1x_3 - 1.4x_2x_3 \quad (2)$$

$$Y_2 = 1,079.3 + 107.0x_1 + 100.1x_2 - 37.3x_3 - 6.6x_1^2 - 2.9x_2^2 - 74.2x_3^2 + 76.3x_1x_2 + 10.5x_1x_3 + 29.0x_2x_3 \quad (3)$$

$$Y_3 = 7.2 - 0.6x1 - 0.3x_2 + 0.1x_3 - 0.4x_1^2 - 0.1x_2^2 - 0.3x_3^2 + 0.04x_1x_2 - 0.1x_1x_3 - 0.3x_2x_3 \quad (4)$$

Eq. (2) (3) and (4) indicated that x_1, x_2 and x_3 showed linear, quadratic, and interaction effects. The three-dimensional responses (Figure 1a, b, c) were predicted values of Y_1 Y_2 and Y_3 based on the range values of x_1, x_2 and x_3.

In Figure 1a, b, the values of Y_1 and Y_2 continuously increased as increasing the ratio of PCF:WF, CS and WC and then gradually decreased as further increase of these variables. Although, Li *et al.* (2011) demonstrated that water content over 21.5% in blue corn cookie could reduce the degradations of anthocyanin and phenolic compounds at high temperature condition. However, decrease of a_w from 0.99 to 0.34 mainly affected to the increase of anthocyanin degradation (Gradinaru *et al.*, 2003). As the results of Zazueta-Morales *et al.* (2002), who reported that in higher temperature and lower a_w conditions could increase the anthocyanin degradation almost 70 times. Jiménez *et al.* (2012) revealed that the anthocyanin degradation of blackberry product above 100°C was significantly affected of maillard reaction which condensed between reducing sugar and amino acids (Erlandson and Wrolstad, 1972). The higher maillard reaction is generated at the intermediate a_w (0.5-0.8) and then slightly declined as increasing a_w value. This might be affected of the reactors in solvent, which are diluted with water following law of mass action (Gradinaru *et al.*, 2003).

ABTS-RSA of PCC had increased as the increase of TAC. As the results of our previous study, we

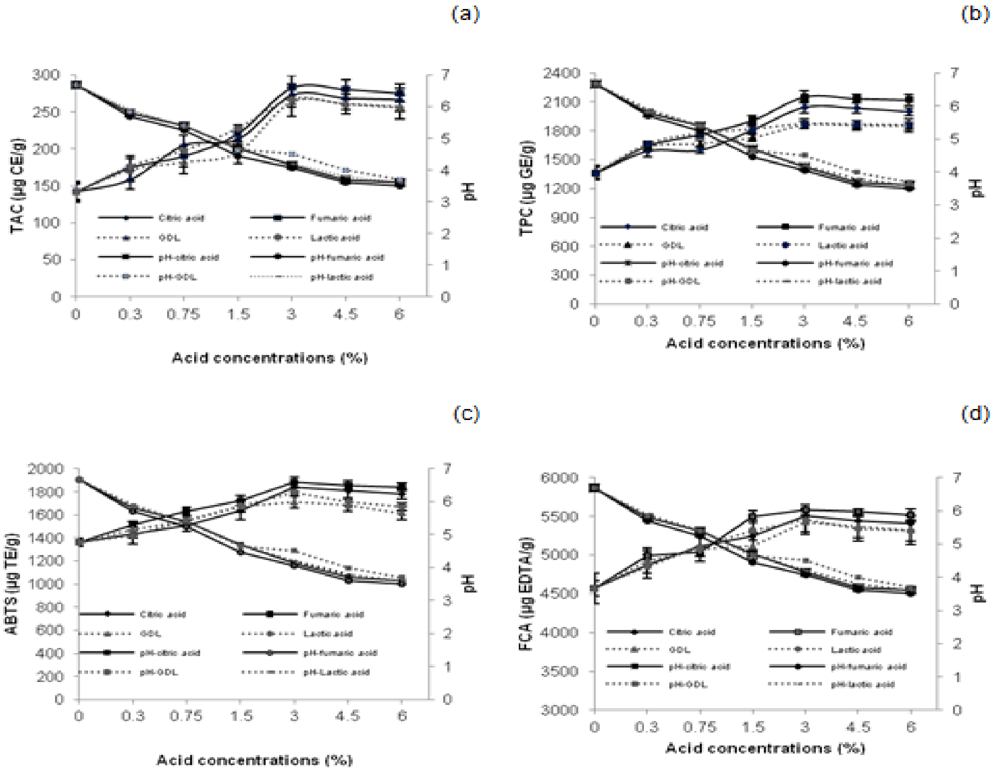

Figure 2. Effect of various levels of citric acid, fumaric acid, GDL and lactic acid on (a) total anthocyanin content (TAC), (b) total phenolic content (TPC), (c) ABTS radical scavenging activity (ABTS-RSA) and (d) ferrous chelating ability (FCA) and pH in purple corn cakes containing corn silk

found that the antioxidant activities of purple waxy corn cookie depended on amount of anthocyanin content (Kokkaew *et al.*, 2015). According to the research of Bily *et al.* (2004), who revealed that the anthocyanin compounds of purple waxy corn product affected to its higher antioxidant capacities. Anthocyanin structure composes of high amount of hydroxyl group which strongly exhibited the free radical scavenging (Del Pozo-Insfran *et al*. (2006).

On the other hand, Figure 1c showed that Y_3 decreased as increase of the ratio of PCF:RF and CS. The texture score of the PCC inversely trend with TAC and ABTS-RSA (Figure 1a,b,c). The variation of PCF:RF and CS in formula might significantly affect to the alteration of PCC texture. As the results of Arafa *et al.* (2012), who reported that the texture of cake was decreased as increasing the corn silk powder content.

Three verification experiments were conducted to confirm the validity of the statistical model (data not shown). The results showed that all observation values were not significant different from the predicted values ($p<0.5$). Therefore, the statistical model was satisfying and useful for maximizing TAC, ABTS-RSA and texture score of PCC. These confirmed that RSM was appropriately used to optimize the PCF and CS for maximal the TAC, ABTS-RSA and texture score of PCC.

Effect of acidulants on TAC and TPC

Figure 2a, b demonstrated that the acidified PCC with citric acid, fumaric acid, glucono-gdelta-lactone (GDL) and lactic acid showed higher of TAC and TPC than nonacidified PCC. The results suggested that adding organic acids in PCC recipes could reduce the TAC and TPC degradation during baking processes. TAC and TPC dramatically increased with the addition of each acid at all levels and the increase also reached plateau at about 3% acid and then slightly decreased. Especially, adding 3% fumaric in PCC showed the highest TAC and TPC (282.7 µg CE/g and 2,154.7 µg GE/g) followed by citric acid (272.4 µg CE/g and 2,045.7 µg GE/g), lactic acid (267.5 µg CE/g and 1,871.3 µg GE/g) and GDL (263.2 µg CE/g and 1,857.7 µg GE/g), respectively. According to the study of Li *et al*. (2011) revealed that adding organic acids in cookie recipe to decrease the pH values could significantly increase TAC in cookies. Del Pozo-Insfran *et al*. (2006) indicated that acidified dough decreased TPC and TAC degradation of tortilla product since organic acids enhanced the stability of phytochemicals. Citric acid, fumaric acid, lactic acid and GDL are all weak organic acids. Their degree or intensity of sourness varies in the decreasing order of fumaric acid, citric, lactic and GDL, respectively

Table 3. Color values and sensory score of purple corn cake containing corn silk powder

Sample	Color			Sensory score			
	Chroma (C^*)	Hue (h)	Color difference (ΔE)	Color	Taste	Texture	Overall acceptability
Purple corn cake	6.9±0.1^{o}	0.6±0.0^{a}	22.8±1.4^{u}	6.3±1.3^{e}	6.8 ±1.1^{a}	6.8 ±1.1^{a}	6.3±1.2^{c}
+Citric acid (0.3% w/w)	7.3±0.5^{m}	0.5±0.1^{e}	23.3±1.4^{t}	6.2±1.4^{e}	6.5±1.2^{a}	7.0±1.2^{a}	7.2±1.0^{a}
+Citric acid (0.75% w/w)	8.0±0.9^{k}	0.5±0.0^{a}	27.2±0.5^{n}	6.8±1.3^{c}	6.3±1.2^{b}	6.8±1.2^{b}	6.8±1.1^{b}
+Citric acid (1.5% w/w)	9.0±0.5^{h}	0.4±0.0^{n}	28.6±0.6^{k}	7.1±0.9^{ab}	6.0±0.8^{c}	6.9±0.8^{a}	6.3±1.0^{c}
+Citric acid (3.0% w/w)	9.7±0.5^{f}	0.4±0.1^{m}	30.8±4.2^{i}	7.4±1.0^{a}	5.8±1.0^{cd}	6.2±1.0^{c}	6.4±1.1^{c}
+Citric acid (4.5% w/w)	9.9±0.5^{e}	0.3±0.0^{r}	31.6±0.4^{g}	7.3±1.2^{ab}	5.5±1.1^{d}	5.8±1.1^{d}	6.0±0.9^{d}
+Citric acid (6.0% w/w)	11.0±0.5^{b}	0.3±0.0^{p}	32.2±0.4^{e}	7.4±1.1^{a}	5.0±1.2^{e}	6.0±1.2^{e}	5.3±1.0^{e}
+Fumaric acid (0.3% w/w)	7.1±0.8^{n}	0.5±0.0^{c}	24.8±0.8^{r}	6.3±1.3^{e}	6.6±1.2^{a}	7.0±1.1^{a}	7.3±1.1^{a}
+Fumaric acid (0.75% w/w)	8.6±0.3^{j}	0.5±0.0^{e}	25.9±0.3^{p}	6.9±1.2^{c}	6.4±1.2^{b}	6.9±1.3^{a}	6.9±1.3^{b}
+Fumaric acid (1.5% w/w)	9.2±0.4^{g}	0.4±0.0^{k}	26.9±0.6^{o}	7.4±0.8^{a}	6.1±0.8^{c}	6.8±0.9^{a}	6.2±0.7^{c}
+Fumaric acid (3.0% w/w)	10.0±1.1^{e}	0.3±0.0^{r}	27.8±0.8^{m}	7.5±1.1^{a}	5.9±1.0^{c}	6.3±1.2^{c}	6.3±1.1c
+Fumaric acid (4.5% w/w)	11.0±0.1^{bc}	0.2±0.0^{t}	28.5±0.6^{k}	7.6±1.2^{a}	5.2±1.1^{d}	5.9±1.1^{d}	5.9±0.9d
+Fumaric acid (6.0% w/w)	12.2±0.6^{a}	0.2±0.0^{q}	29.6±0.7^{i}	7.7±1.2^{a}	5.0±1.2^{e}	6.1±1.2^{c}	5.4±1.0e
+ GDL (0.3% w/w)	7.2±1.4^{n}	0.4±0.1^{i}	25.4±2.7^{q}	6.2±1.3^{e}	6.6±1.2^{a}	7.0±1.3^{a}	7.1±1.1^{a}
+ GDL (0.75% w/w)	7.8±0.3^{l}	0.4±0.0^{gn}	24.3±2.7^{s}	6.3±1.1^{e}	6.1±1.2^{b}	6.9±1.3^{a}	6.7±1.3^{b}
+ GDL (1.5% w/w)	8.0±0.2^{k}	0.4±0.0^{n}	28.1±2.0^{l}	7.2±1.0^{ab}	6.0±0.8^{c}	6.9±0.9^{a}	6.2± 0.8^{c}
+ GDL (3.0% w/w)	9.4±0.9^{g}	0.3±0.1^{e}	29.4±0.6^{j}	7.3±1.1^{ab}	5.9±1.0^{c}	6.1±1.2^{c}	6.4±1.1^{c}
+ GDL (4.5% w/w)	10.1±0.3^{e}	0.3±0.0^{p}	31.1±0.8^{i}	7.4±1.2^{ab}	5.4±1.1^{d}	5.9±1.1^{d}	6.1±0.7^{d}
+ GDL (6.0% w/w)	10.8±0.3^{d}	0.2±0.0^{q}	34.0±0.3^{c}	7.6±1.3^{a}	4.8±1.2^{e}	6.1±1.2^{c}	5.2±1.0^{e}
+Lactic acid (0.3% w/w)	7.1±0.5^{n}	0.4±0.0^{g}	31.3±1.6^{h}	6.1±1.1^{e}	6.4±1.2^{b}	7.0±1.2^{a}	7.0±1.1^{a}
+Lactic acid (0.75% w/w)	7.7±0.6^{l}	0.4±0.0^{i}	31.7±0.5^{g}	6.2±1.2^{e}	6.0±1.2^{c}	6.9±1.3^{a}	6.9±1.1^{b}
+Lactic acid (1.5% w/w)	7.8±0.1^{l}	0.4±0.0^{m}	32.2±1.8^{f}	7.3±0.7^{ab}	5.9±0.8^{c}	6.5±1.0^{b}	6.4± 0.8^{c}
+Lactic acid (3.0% w/w)	8.8±0.6^{i}	0.3±0.0^{o}	33.2±1.1^{e}	7.3±1.1^{ab}	5.7±1.0^{c}	6.3±0.9^{bc}	6.5±1.1^{c}
+Lactic acid (4.5% w/w)	9.4±0.6^{g}	0.3±0.0^{p}	34.4±0.6^{b}	7.4±1.2^{a}	5.5±1.1^{d}	6.4±1.2^{b}	6.1±0.9^{d}
+Lactic acid (6.0% w/w)	10.9±1.3^{cd}	0.2±0.0^{q}	35.2±0.8^{a}	7.5±1.3^{a}	4.9±1.2^{e}	6.1±1.1^{d}	5.4±1.0^{e}

*Values are expressed as means + standard deviation
*The different letters in the same column are significantly different ($p<0.05$)

at equal concentration (Watine, 1995). Fumaric acid is the strongest organic food acid among this four acids. The sour taste response imparted to a food is attributed to the hydrogen (H^+) or hydronium (H_3O^+) ions (Berry, 2001). In addition, the more free anions associated with an acid and the more anthocyanin retention in the lower pH cakes (Francis, 1999).

Effect of acidulants on Antioxidant activities

ABTS-RSA and FCA increased as increasing of all acid concentrations. Acidified PCC with 3% organic acids showed the highest ABTS-RSA and FCA (Figure 2c, d). However, acidified PCC with 3% fumaric acid demonstrated the highest the ABTS-RSA and FCA (1,884.8 µg TE/g and 5,581.1 µg EDTA/g) followed citric acid (1,840.3 µg TE/g and 5,512.1 µg EDTA/g), lactic acid (1,795.1 µg TE/g and 5,453.7 µg EDTA/g) and GDL (1,712.1 µg TE/g and 5,430.6 µg EDTA/g), respectively. The results showed that antioxidant activities of PCC depended on TAC and TPC. According to the research of Chang *et al.* (2002) reported that antioxidant properties of purple corn depended on phenolic content especially feruric acid and p-coumaric acid. Moreover, Bily *et al.* (2004) revealed that the anthocyanin or phenolic compounds affected to the higher antioxidant capacities of purple corn product. These phenolic compounds are agents exhibiting reducing, donating hydrogen and singlet oxygen scavenging properties meanwhile flavonoid compounds are composed of hydroxyl group that exhibited the free radical scavenging and ferrous chelating capacities (Del Pozo-Insfran *et al.*, 2006). From the results, adding organic acids affected to the higher antioxidant properties of purple corn products due to their reducing activity (Li *et al.*, 2011).

Effect of acidulants on color values

The changes in TAC retention were evaluated the instrumental color values (Table 3). The brown pigment formations are a clarification for the colorimetric changes during high temperature process (Jiménez *et al.*, 2012). The acidified PCC (APCC) with 4 organic acids exhibited significantly higher the difference color (ΔE) values than nonacidified PCC ($p<0.5$). Additionally, ΔE values of all APCC were increased as increase of acid concentrations (Table 3). Acidification in PCC induced the increase of product brightness due to hydrogen peroxide produced from acid oxidation or condensation between acids and anthocyanins (Poei-Langston and Wrolstad, 1981).

Hue (h) values of APCC were significantly lower than non-APCC. Increase of acid concentration in PCC (decreasing pH) exhibited a decrease in hue values (Table 3). The lower pH of APPC favored a purple hue more than the burgundy-brown. Meanwhile, non-APCC might be appeared to the human eye as deeper purple color. The lower pH could induce the oxidation of anthocyanin affected to a bleaching of purple color (Buttery and Ling, 1995). Therefore, the change of hue value of PCC could be influence of reduction of anthocyanin content due to high temperature. According to the results of Poei-Langston and Wrolstad (1981), who revealed that the red color of corn purple cookie product was increased with adding 4% ascorbic solution.

The changes in chroma (C^*) values were shown in Table 3. C^* values increased as increase of acid concentration indicating a decrease in the opaqueness of PCC surface. These might be cause by changes in the visual characteristics due to furfural organic compounds. Furfural and its derivatives from maillard reaction can react with anthocyanins to form the colorless and dark compounds (Es-Safi *et al*., 2000). Organic acids, which are reducing agents, significantly improve the color of products (Berry, 2001). In addition, organic acids significantly enhanced the reduction of the anthocyanin degradation by a condensation mechanism and polymeric pigment formation (Chang *et al*., 2002). However, adding organic acids in higher concentration could enhance the maillard reaction affected to change of red or purple of anthocyanins to brown pigment (Chaovanalikit *et al*., 2006). In our study, the higher level of acid concentration in PCC showed pinker rather than brown, which indicated more remaining anthocyanin content after baking. Notably, adding 3% organic acids in PCC demonstrated the highest TAC and TPC as well as producing the desirable color appearance of PCC.

Effect of acidulants on sensory evaluation

Table 3 showed the sensory score for color, taste, texture and overall acceptability of APCC. The results revealed that APCC with 0.3% acids exhibited the highest score of taste, texture and overall acceptability and then significantly decreased as increasing acid concentrations. Meanwhile, the color score were increased as the results of increase of acid concentrations. However, the color scores of APCC were inversely trend with the taste, texture and overall acceptability score. Therefore, higher acid concentration in PCC, which exhibited the higher of TAC, TPC, ABTS-RSA, FCA and color values, was not desirable the acceptance of panelists on the products.

Conclusion

The optimal condition to maximize the TAC (142.0 µg CE/g), ABTS-RSA (1,249.0 µg TE/g) and texture score of PCC (6.6) was PCF:RF of 71.1:28.9, CS of 10.0 and WC of 27.1 (% total flour weight basis). APCC with 3% fumaric acid exhibited the highest reduction of TAC, TPC, ABTS-RSA, TPC degradation as well as producing the desirable color appearance. Notably, the higher acid concentration in PCC showed the lower sensory score in taste, texture, and acceptability. Additionally, texture analysis will further study to investigate the effects of acidulants on PCC qualities. This study would be alternative way for sustainable management and added values of agricultural materials to prepare the functional foods with high nutrition and health benefit.

Acknowledgement

We are grateful to the Nakhon Phanom University for financial support.

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