

Optimization of extrusion conditions for ready-to-eat breakfast cereal enhanced with defatted rice bran

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Abstract

Defatted rice bran was incorporated with cereal base by an extrusion process to obtain a functional, ready-to-eat breakfast cereal. This study investigated the effects of defatted rice bran content (10, 15 and 20%), the sixth barrel temperature (H6: 130, 140 and 150°C), and feed moisture content (14, 17 and 20%) on the physical and functional properties of extrudates in breakfast cereal production using response surface methodology (RSM) and Box-Behnken experimental design. The results showed high correlation coefficients (R^2) of multiple regression equations about the relationship among product responses and process variables, and no significant lack of fit which indicating a best-fit model. The optimum conditions were determined from the overlaid area found by superimposing the individual contour plots of responses. Correspondingly, the verification of results showed good agreement between the responses of experimental values at selected optimum conditions and the predicted values from regression equations.

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Introduction

Defatted rice bran, a low valued co-product of rice milling and rice bran oil extraction, has great potential as a supplementary source of many nutrients. After extraction of the edible oil from rice bran, defatted rice bran is remained and used in animal feed or is discarded as agricultural waste. However, defatted rice bran by-products have unique functional and nutritional properties. It still contains significant amounts of various kinds of protein, starch, polysaccharides, vitamins, minerals, dietary fiber, and phenolic substances (Wiboonsirikul *et al.*, 2007), which are beneficial as health promoting and functional substances in foods. Research studies have shown that soluble rice bran fiber extracted from defatted rice bran is known for its antioxidant, anti-tumour activities and hypocholesterolemic effects in human (Aoe *et al.*, 1993; Abdul and Yu, 2000; Tsutsumi, 2000; Fabian *et al.*, 2010). In comparison with raw bran, defatted rice bran can be preserved and can be stored for a long period of time without rapid deterioration of crude fat as occurring in the raw rice bran which produce an off-flavor and soapy taste unfit for human consumption (Zullaikah *et al.*, 2005). These confirm that the defatted rice bran has great potential in food applications, especially in development of functional foods.

Although the health benefits of defatted rice bran

have been recognized, it is still underutilized and sold as an ingredient for the pet food industry. According to the previous researches, very few studies have been published concerning the incorporation of defatted rice bran into processed foods such as bread sticks and fried batter coating. It might be of value to develop special nutritious food products from bran and provide marketing opportunity for incorporation of defatted rice bran into extruded products which have numerous advantages in manufacturing over conventional processing methods, such as faster processing times, lower processing costs, less square footage required, shorter response time, and greater flexibility (Riaz, 2000). Extruded breakfast cereal is one of ready-to-eat extruded products and ideal food for people's modern-day lifestyle, where speed and convenience, as well as complete nutritional values, are desirable food characteristics. Compared to other commercial breakfast cereal products, the utilization of defatted rice bran in the manufacture of extruded breakfast cereal has not yet available in the market. Relatively, little research has been conducted on how non-starch polysaccharides behave in extrusion processes or the potential effect of implementing defatted rice bran into extruded breakfast cereal in relation to the physicochemical and functional properties of ready-to-eat breakfast cereal products.

Therefore, the present study was designed to utilize rice industrial by-product for value

addition. Response surface methodology (RSM), an empirical modeling technique, was applied to optimize production of ready-to-eat breakfast cereal enhanced with defatted rice bran, and to investigate the relationship of controllable operating conditions affecting the cereal's physical and functional properties. The optimization process of extrusion conditions and the verification of results found in this research will determine the optimum levels of defatted rice bran, barrel temperature and feed moisture content for producing an extruded breakfast cereal with increased total dietary fiber, total phenolic compounds and antioxidant activity, while still providing good physical properties and acceptable sensory quality.

Materials and Methods

Preparation of raw materials

The ingredients used for extruded ready-to-eat breakfast cereal preparation consisted of the composition (%) between rice flour and defatted rice bran equaled to 50 : 10, 45 : 15 or 40 : 20, while the other ingredients were fixed at 20% corn grit, 4.5% defatted soy flour, 8% full-fat soy flour, 5% modified starch, 1% rice bran oil, 1% premixed vitamins, and 0.5% calcium carbonate. Defatted rice bran was obtained from the polishing process that produces white rice, after removing oil from extraction process, grinding and cleaning, which was supplied by Patum Rice Mill and Granary Public Co., Ltd. (Pathum Thani, Thailand). Other ingredient, such as modified starch (Instant Textaid A) was supplied by National Starch and Chemical Limited (Samut Prakan, Thailand).

Extrusion process

Extrusion was performed in a co-rotating twin screw extruder ZE25 (KraussMaffei Berstorff GmbH, Hanover, Germany) which consisted of 7 barrel parts ending with a 24.5 mm thick die plate and one floral-shaped die hole. The floral-shaped die hole in this experiment made the product look like flower which similar to circular shape with 7 mm in diameter. The length to diameter (L/D) ratio of the extruder was 870/25. A standard design of screw configuration for processing cereals and flour-based products was used. Mixtures of raw materials with varying defatted rice bran content (10, 15 and 20%) were fed into the extruder by a volumetric twin screw feeder. Water was pumped into the ingredients to achieve the required moisture content. The blends were extruded at varying temperature profiles or the sixth barrel temperature (H6: 130, 140 and 150°C) and

Table 1. Box-behnken design for experimental runs with different combination of extrusion variables

Run No.	Coded Values (Actual Values)		
	Defatted rice bran content (%)	Temperature at the sixth barrel: H6 (°C)	Feed Moisture (%)
1	0 (15)	-1 (130)	-1 (14)
2	0 (15)	+1 (150)	-1 (14)
3	0 (15)	-1 (130)	+1 (20)
4	0 (15)	+1 (150)	+1 (20)
5	-1 (10)	-1 (130)	0 (17)
6	-1 (10)	+1 (150)	0 (17)
7	+1 (20)	-1 (130)	0 (17)
8	+1 (20)	+1 (150)	0 (17)
9	-1 (10)	0 (140)	-1 (14)
10	-1 (10)	0 (140)	+1 (20)
11	+1 (20)	0 (140)	-1 (14)
12	+1 (20)	0 (140)	+1 (20)
13	0 (15)	0 (140)	0 (17)
14	0 (15)	0 (140)	0 (17)
15	0 (15)	0 (140)	0 (17)

feed moisture content (14, 17 and 20%). After stable conditions were established, extrudates were collected and dried in an air oven at 80°C for 15 min. The extrudates at varying conditions were coated in white chocolate syrup to obtain the finished products and then examined for physical and functional properties in order to optimize the operating conditions.

Experimental design and statistical analysis

A Box-Behnken design of response surface methodology (RSM) was employed, with three independent variables and three levels of each variation. This comprised 15 experimental runs, of which there were center points, as shown in Table 1. The independent variables were defatted rice bran content (10, 15 and 20%), the sixth barrel temperature (H6: 130, 140 and 150°C), and feed moisture content (14, 17 and 20%). Range of these variables was established by preliminary tests. In this study, data obtained by application of RSM was used to build mathematic models to interpret the relationship between the independent and dependent variables. Dependent variables were physical properties (expansion ratio, bulk density, water absorption index, water solubility index and texture measurement) and functional properties (dietary fiber content, total phenolic compounds and antioxidant activity) of extrudates. Regression analysis was performed based on the experimental data and was fitted to an empirical second-order polynomial model, as shown in the following equation:

$$Y = \beta_0 + \sum_{i=1}^3 \beta_i X_i + \sum_{i=1}^3 \beta_{ii} X_i^2 + \sum_{i=1}^2 \sum_{j=i+1}^3 \beta_{ij} X_i X_j + e$$

where Y was the response variable; β_0 , β_i , β_{ii} , β_{ij} , were the regression coefficients of variables for intercept, linear, quadratic and interaction terms, respectively; and x_i and x_j were independent variables. The results of the experimental design were analyzed and interpreted using MINITAB version 15 statistical software package. Surface plots were also generated as a function of two variables, while holding the value of a third variable constant (at the central value).

Optimization and verification

For optimization, statistical calculations were used to generate overlaid contour maps from the regression models which gave no significant lack-of-fit and high R^2 . A proper choice of optimum conditions is desirable to achieve a good appearance of the physical product and improved functional properties; this can be determined from the overlaid graph of the response contour plots. Finally, verification experiments were carried out to confirm the adequacy of the models for predicting the optimum operating conditions. Good agreement between the experimental results obtained under the selected conditions from RSM optimization and the predicted values indicated verification of the results.

Product analysis

Expansion ratio

To determine the expansion ratio, the cross-sectional diameter of the extrudate was measured with a vernier caliper. The expansion ratio was calculated as the cross-sectional diameter of the extrudate divided by the cross-sectional diameter of the die opening (Ding *et al.*, 2005). The expansion ratio values were obtained from 10 random samples for each extrusion condition.

Bulk density

The extrudates at each extrusion condition were poured into a 100 ml measuring cylinder and tapped ten times on a flat wooden platform. The volume and weight of the extrudates were recorded to calculate the bulk density by unit mass per unit volume (Shittu and Lawal, 2007).

Water absorption index (WAI) and water solubility index (WSI)

The WAI and WSI were measured using a technique developed for cereals (Anderson *et al.*, 1969; Ding *et al.*, 2006). Ground extrudates were suspended in water at room temperature for 30 min, gently stirred during this period, and then centrifuged at 3000xg for 15 min. The supernatant was decanted into an evaporation dish of known weight. The WAI is the weight of the gel obtained after removal of the supernatant per unit weight of original dry solids. The WSI is the weight of dry solids in the supernatant expressed as a percentage of the original weight of the sample.

Textural measurement

The textural characteristics of extrudates were measured using a TA-XT2i texture analyzer (Stable

Micro Systems, Surrey, UK) with a 2 mm cylindrical probe (P/2). The instrument was fitted with the standard 5 kg load cell supplied with the texture analyzer. The extrudates were punctured by the probe at a speed of 2.0 mm/s to a distance corresponding to approximately 30% of the height of the extrudates. A force-time curve was recorded to calculate the maximum peak force, which represents the resistance of the extrudate to penetration (the hardness of the extrudate). Ten randomly collected samples of each extrudate at varying operating conditions were measured, and a mean taken.

Dietary fiber determination

Total dietary fiber (TDF), insoluble dietary fiber (IDF) and soluble dietary fiber (SDF) were determined according to the enzymatic-gravimetric (Phosphate buffer) AOAC methods (985.29, 991.42 and 993.19). Briefly, sample duplicates (1 g of each duplicate powdered sample) were homogenized in phosphate buffer (pH 6.0) and incubated at 100°C with a heat-stable α -amylase under constant agitation and then at 60°C treated with a protease and amyloglucosidase to remove protein and starch, respectively. To determine the TDF content, the enzyme-digested samples were treated with 95% (v/v) ethanol (ethanol/sample ratio, 4:1, v/v) at room temperature for 1 h to precipitate soluble fiber. The residues were filtered on fritted crucible and washed sequentially with 78% (v/v) ethanol, 95% (v/v) ethanol and absolute acetone, and dried overnight at 105°C.

To determine IDF and SDF content, the enzyme-digested sample was filtered and the insoluble material washed twice with preheated water at 60°C and then treated as above to give IDF. Four volume of 95% (v/v) ethanol were then added to the filtrate and placed at room temperature for 1 h. After filtration, the recovered precipitate, SDF, was dried at 105°C. TDF, IDF and SDF were corrected for residual protein (Kjeldhal method) and ash (525°C, 5 h).

Total phenolic compounds

The Folin-Ciocalteu method was employed, as described by Li *et al.* (2007), with some modifications for extruded products. One ml of Folin-Ciocalteu reagent (diluted 1:10) was added to the methanolic extract of each sample (0.2 ml). After 4 min, 0.8 ml of saturated Na_2CO_3 solution was added. After 30 min of incubation at room temperature, the sample tube was centrifuged for 10 min at 5,000 rpm. Absorbance of supernatant was measured at 765 nm. Gallic acid was used for calibration of the standard curve. The results were expressed as mg gallic acid equivalent (mg GAE)/g weight of sample.

Antioxidant activity

This test was performed using a stable radical, DPPH, as described by Tachibana *et al.* (2001), with some modifications. In brief, antioxidant activity was determined by the reaction of methanolic extract of each sample (3 ml) and 200 μ M DPPH (3 ml). Absorbance of the samples was measured at 515 nm after 40 min incubation at room temperature in the dark. A calibration curve was made using Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid) – a synthetic, hydrophilic vitamin E analog – as an external standard, with a range of concentrations from 0 to 100 μ M. Results were expressed as Trolox equivalents (Sensoy *et al.*, 2006).

Results and Discussion

Effects of extrusion conditions on the physical and functional properties of extrudates were investigated; the results of regression analysis in terms of coded variables are shown in Table 2. The regression models for product responses had high coefficient of determination ($R^2 > 0.8$) and none showed significant lack of fit ($P > 0.05$), indicating that all the second-order polynomial models correlated well with the measured data. These can be used to estimate the relationship between the operating conditions and product properties, and can then be applied to optimize the preparation process of ready-to-eat breakfast cereal enhanced with defatted rice bran.

Product responses

Expansion ratio and Bulk density

Expansion ratio and bulk density, which are important characteristics for extruded puffed products, can be controlled by the configuration of extrusion operating conditions, including raw material specifications. Bulk density is an index of puffing, in which extrudates having lower expansion show higher density and vice versa. In this research, an inverse relationship was found between expansion ratio and density of extrudates, as in an earlier report (Singh *et al.*, 1996; Ding *et al.*, 2005; Thymi *et al.*, 2005; Ding *et al.*, 2006). Regression analysis results (Table 2) showed that all of the variables – defatted rice bran content, barrel temperature and feed moisture content – had a significant effect on expansion ratio and bulk density. Increased defatted rice bran content (Figure 1a, 1b, 2a and 2b) resulted in an extrudate with lower expansion and higher bulk density. Fiber from defatted rice bran has a significant effect on product structure and texture:

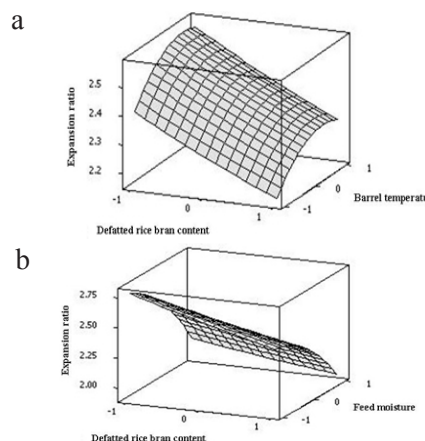


Figure 1. Response surface plot displaying the effect of operating conditions on the expansion ratio of extrudate as a function of (a) defatted rice bran content and barrel temperature: H6 and (b) defatted rice bran content and feed moisture. (Measured at the central point of the third variable.)

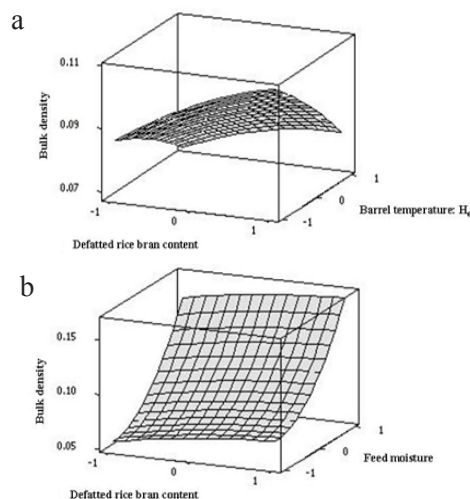


Figure 2. Response surface plot displaying the effect of operating conditions on the bulk density of extrudate as a function of (a) defatted rice bran content and barrel temperature: H6 and (b) defatted rice bran content and feed moisture. (Measured at the central point of the third variable.)

it does not expand but acts as a solid filler, diluting the expanding starch ingredient in the mix. The bulk density of the product thus increases with increasing fiber content (Ryu, 2004). Feed moisture has also been found to affect product density. Increased feed moisture content during extrusion decreased the expansion ratio and increased the bulk density (Figure 1b and 2b). The high dependence of bulk density and expansion on feed moisture would reflect its influence on the elasticity of the starch-based material. Thymi *et al.* (2005) reported that increasing the moisture content during extrusion changed the amylopectin molecular structure of the starch-based material, reducing the melt elasticity (resulting in reduced gelatinization), decreasing the expansion and

Table 2. Regression coefficients of the polynomial function and the coefficients of determination (R^2) calculated on coded values

Variables	Expansion ratio	Bulk density (g/ml)	WAI	WSI (%)	Texture:Maximum force (g)	Dietary fiber			Total phenolic compound (mg GAE/100 g)	Antioxidant activity (mmol Trolox/100 g)
						IDF (%)	SDF (%)	TDF (%)		
Intercept	2.41667**	0.090400**	4.10273**	28.9217**	1528.24**	6.33297**	1.92883**	8.24927**	132.761**	7.04270**
x_1	-0.12500**	0.008375**	-0.12419**	-2.5318**	171.69**	1.51678**	0.07799	1.58008**	14.429**	1.34084**
x_2	0.07000*	-0.010975**	0.07139*	0.5421	-22.61	0.15169	0.23408	0.37897	3.304	0.14581
x_3	-0.29500**	0.045800**	0.11855**	-2.4699**	222.78**	-0.08724	-0.12254	-0.21577	-11.483**	-1.19483**
x_1x_1	0.00917	-0.002825	0.02617	-0.1156	-31.63	0.04218	-0.08309	-0.06665	-4.325	0.06764
x_2x_2	-0.06583	-0.001625	-0.16578*	0.8288	19.85	-0.60480*	0.48678	-0.09385	-3.680	-0.49861
x_3x_3	-0.11083**	0.022825**	-0.16575**	0.4560	229.89**	0.03380	-0.28334	-0.22970	-4.136	-0.12499
x_1x_2	-0.00500	-0.002850	0.07792	-0.7282	5.52	0.00885	0.33917	0.36003	0.390	-0.05593
x_1x_3	0.04000	-0.000100	-0.06760	0.9185	90.30*	-0.48835*	0.21520	-0.29157	-2.748	-0.93230
x_2x_3	0.03000	-0.009600**	0.07380*	0.5684	-19.62	-0.38122	-0.11832	-0.50112	-0.236	-0.14900
R square (%)	98.6	99.7	97.4	95.2	97.8	97.6	79.4	95.7	95.3	97.7
Sig. F < 0.05	0.052	0.082	0.053	0.093	0.064	0.052	0.308	0.341	0.236	0.062

* Significant at the 5% level *

** Significant at the 1% level **

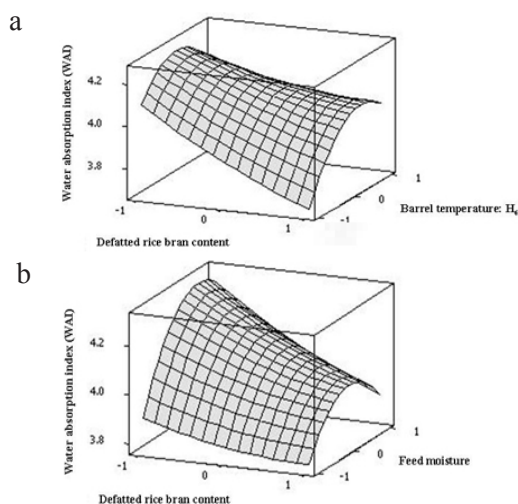
 x_1 = Defatted rice bran content (%), x_2 = Temperature at the sixth barrel: H6 (°C), x_3 = Feed moisture (%)

Figure 3. Response surface plot displaying the effect of operating conditions on the water absorption index (WAI) of extrudate as a function of (a) defatted rice bran content and barrel temperature: H6 and (b) defatted rice bran content and feed moisture. (Measured at the central point of the third variable.)

increasing the density of the extrudates. Likewise, increased barrel temperature (Figure 1a and 2a) resulted in an extrudate with higher expansion and lower bulk density. At high temperature, there was sufficient heat to produce steam and therefore drive expansion, so expansion was high leading to reduce density. Other studies have observed similar trends (Barrett and Peleg, 1992; Pan *et al.*, 1998; Ding *et al.*, 2005; Ding *et al.*, 2006; Stojceska *et al.*, 2009).

Water absorption index (WAI) and Water solubility index (WSI)

When extruded starches are dispersed in an excess of water, their main functional properties are water absorption and water solubility. Native starch does not absorb water at room temperature, and its viscosity is nearly zero, whereas extruded starch absorbs water rapidly to form a paste or gel at room temperature. WAI correlates well with cold-paste viscosity because only damaged starch granules absorb water at room temperature and swell, creating increased viscosity. In this research, WAI decreased

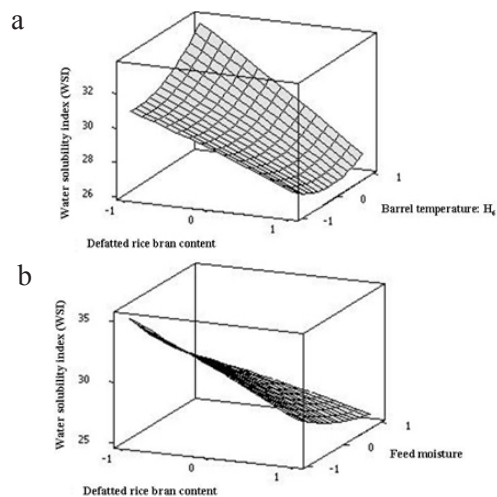


Figure 4. Response surface plot displaying the effect of operating conditions on the water solubility index (WSI) of extrudate as a function of (a) defatted rice bran content and barrel temperature: H6 and (b) defatted rice bran content and feed moisture. (Measured at the central point of the third variable.)

with higher rice bran content as shown in Figure 3a and 3b. The results indicated that the presence of more bran in the mixture reduced the availability for gelatinization of the starch granules, thus reduced viscosity and WAI because of the replacement of the starch by fiber component. In contrast, reducing of bran content in the mixture caused more open structure of starch granules for allowing water penetration and retention to get more gelatinization and higher WAI. The similar effect of bran content on structural of starch component has been observed (Grenus *et al.*, 1993; Hashimoto and Grossmann, 2003). At higher level of feed moisture as observed in this research, the result has trend to lower WAI as shown in Figure 3b. Probably during extrusion under higher feed moisture content, the vapor pressure is lower resulting from lubricating the screws, which would reduce shear and friction to lower flashing of moisture, and finally in unexpanded structure. This would lower the capacity of water absorption or WAI as the similar results according to Ilo *et al.* (1996).

The water solubility index (WSI) expresses

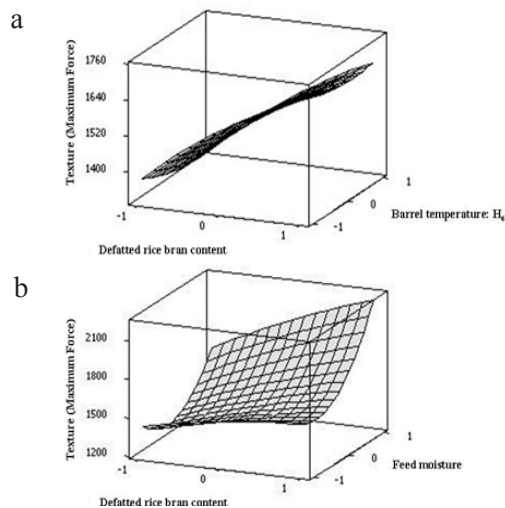


Figure 5. Response surface plot displaying the effect of operating conditions on the hardness (maximum force) of extrudate as a function of (a) defatted rice bran content and barrel temperature: H6 and (b) defatted rice bran content and feed moisture. (Measured at the central point of the third variable.)

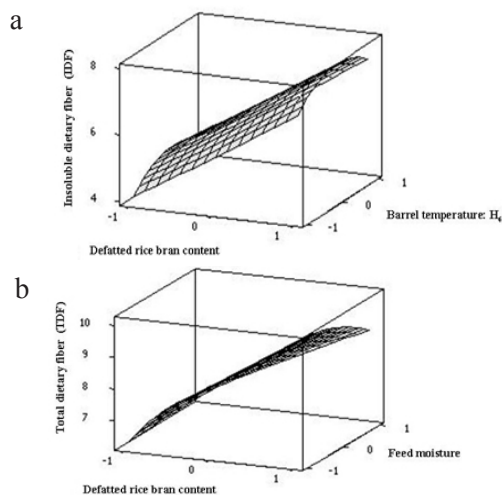


Figure 6. Response surface plot displaying the effect of operating conditions on (a) the insoluble dietary fiber content as a function of defatted rice bran content and barrel temperature: H6 and (b) the total dietary fiber content as a function of defatted rice bran content and feed moisture. (Measured at the central point of the third variable.)

the percentage of dry matter recovered after the supernatant is evaporated from the water absorption determination. WSI is often used as an indicator of degradation of molecular components (Kirby *et al.*, 1988) by measuring the quantity of soluble molecules as related to dextrinization. It was observed in this research that defatted rice bran and feed moisture content had a strong effect on WSI. A decrease in defatted rice bran content resulted in an extrudate with higher water solubility as shown in Figure 4a and 4b, due to more expanded structure increasing the water solubility of starch. A

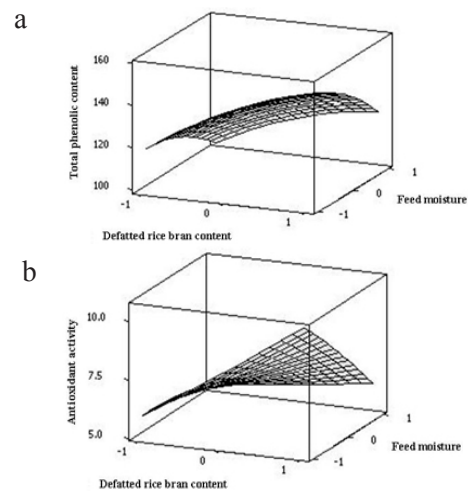


Figure 7. Response surface plot displaying the effect of operating conditions on (a) the total phenolic content and (b) antioxidant activity of extrudates as a function of defatted rice bran content and feed moisture. (Measured at the central point of the third variable.)

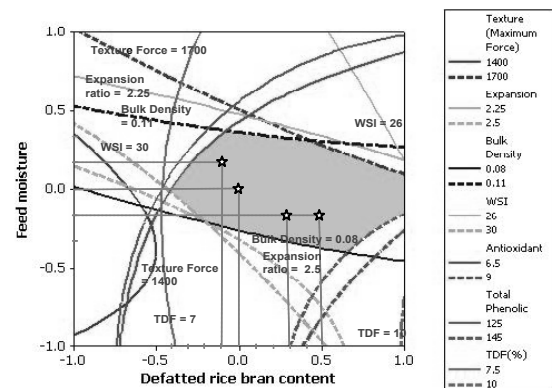


Figure 8. Optimum region identified by the overlaid contour plots of the four responses: expansion ratio, bulk density, WSI, hardness, total dietary fiber, total phenolic compounds and antioxidant activity at different levels of defatted rice bran content and feed moisture while holding optimal barrel temperature.

decrease in feed moisture content also resulted in an extrudate with higher water solubility (Figure 4b), because higher solubility occurred with simultaneous high thermal and mechanical energy inputs which increasing degradation of starch. These observations corroborated the reported earlier (Brennan *et al.*, 2008; Hagenimana *et al.*, 2006; Singh *et al.*, 2007)

Textural measurement

Textural properties of extruded products are important and closely related to the consumer acceptance. The characteristic of extruded ready-to-eat breakfast cereal products should be created with harder texture than snack food, which hydrate more slowly and retain their desired crispness longer when consumed with milk. The hardness and crispness of expanded extrudate is associated with the expansion and cell structure of the product. The instrumental

Table 3. Experimental and predicted values of verification experiment under optimum conditions

Rice bran (%)	Optimum conditions		Expansion Ratio		Bulk Density		WSI		Hardness	
	Temperature : H ₂ (C)	Feed moisture (%)	Predicted value	Real value	Predicted value	Real value	Predicted value	Real value	Predicted value	Real value
-0.10 (14.5)	0 (140)	0.33(17.5)	2.32	2.32 ^{NS} ±0.32	0.11	0.11 ^{NS} ±0.03	28.38	27.92 ^{NS} ±0.74	1606.33	1591.80 ^{NS} ±47.43
0 (15)	0 (140)	0 (17)	2.42	2.42 ^{NS} ±0.03	0.09	0.09 ^{NS} ±0.00	28.92	27.99 ^{NS} ±0.57	1528.24	1549.00 ^{NS} ±31.53
0.30(16.5)	0 (140)	0.33(16.5)	2.46	2.45 ^{NS} ±0.02	0.08	0.08 ^{NS} ±0.00	28.93	28.28 ^{NS} ±0.30	1519.48	1543.60 ^{NS} ±35.40
0.50(17.5)	0 (140)	0.33(16.5)	2.44	2.44 ^{NS} ±0.03	0.08	0.08 ^{NS} ±0.00	28.34	26.36 ^{NS} ±0.81	1542.80	1558.90 ^{NS} ±35.43

^{NS} Not significant for t-test at 95% level confidence.

method for the measurement of hardness is the maximum force required for a probe to penetrate the extrudate. With regard to the hardness of the extrudates, increasing defatted rice bran and feed moisture content resulted in a significant increase in hardness as shown in Figure 5a and 5b. Increasing dietary fiber content from defatted rice bran resulted in rupture of gas cells, which reduced overall expansion and increased hardness. The change of hardness with fiber was probably the result of the effect of this material on the cell wall thickness to make lesser porous, thicker cell walls and harder the extrudates. Similar findings were reported by Mendonca *et al.* (2000); Yanniotis *et al.* (2007) and Ainsworth *et al.* (2007). Likewise, increasing the moisture content possibly resulted in a lower degree of starch gelatinization and lower expansion which showed increasing hardness of extrudates. Previous studies also reported that the hardness of extrudate increased as the feed moisture increased (Liu *et al.*, 2000; Ding *et al.*, 2006; Stojceska *et al.*, 2009).

Dietary fiber

Defatted rice bran, as a functional ingredient gives interesting health benefits with very low allergenic property and a good fiber source which divided into two fractions, soluble and insoluble in water. Soluble dietary fiber is involved in lowering effects on blood cholesterol and glucose intestinal absorption whereas insoluble dietary fiber is mainly related to intestinal regulation, including an increase in fecal bulk, reduced transit time of fecal material through the large intestine and other benefits (Repo-Carrasco-Valencia *et al.*, 2009). The earlier report found that the properties of dietary are affected by food processing. Chang and Morris (1990) found that the heat treatment (autoclave) reduced the content of total and insoluble dietary fiber in apple and oat bran, whereas in case of corn, heat treatment (autoclave and microwave) increased the content of soluble dietary fiber. Some of previous research (Gualberto *et al.*, 1997; Repo-Carrasco-Valencia *et al.*, 2009) found a decrease in the content of insoluble dietary fiber during extrusion cooking and an increase in the content of soluble fiber which resulting from shear stress caused by the high screw speed. However, the effect of barrel temperature and feed moisture ranges in this research were not significant on dietary

fiber, probably due to different varieties of fiber source and less shear stress to breakdown the fiber macromolecules. The observation was only significant on defatted rice bran content as shown in Figure 6a and 6b that higher defatted rice bran content in the recipe resulted in higher insoluble and total dietary fiber of extrudates.

Total phenolic compound and antioxidant activity

Besides of dietary fiber, defatted rice bran has also contained antioxidant compounds which are mainly due to phenolic compounds. These antioxidant compounds may be concentrated in the bran fractions which are associated with the outer layers, particularly the aleurone layer (Miller *et al.*, 2000; Martiane-Tomea *et al.*, 2004). Correspondingly, incorporation of defatted rice bran as a good source of such phytochemicals can enhance the antioxidant activity of ready-to-eat breakfast cereal products. Analysis of extrusion operating conditions showed that the level of defatted rice bran content and feed moisture had a highly significant effect ($P \leq 0.01$) on these functional properties (Figure 7a and 7b). Due to the antioxidant compounds in the bran fractions, increasing of defatted rice bran content resulted in higher total phenolic compounds and antioxidant activity (Figure 7a and 7b). Furthermore, this study found that increasing the feed moisture content resulted in a lower amount of total phenolic compounds and antioxidant activity (Figure 7a and 7b). There are opposing reports of total phenolic compounds and antioxidant activity stability during extrusion process. Some research showed that the extrusion process affected the content of total phenolic compounds and antioxidant activity, decreasing these values (Repo-Carrasco-Valencia, 2009), whereas another research showed that extrusion process (170°C) did not cause any change in antioxidant activity (Sensoy *et al.*, 2006). Stojceska *et al.* (2009) had reported that water feed rate and extrusion cooking had no effect on the level of total phenolic compounds and antioxidant activity in samples containing brewer's spent grain but showed different trend in samples containing red cabbage. These different results could be explained by having different type of phenolic compounds which different effective on antioxidant activities. However, the results suggest that processing conditions can be optimized to keep functionality of active compounds

in ready-to-eat breakfast cereal products.

Optimization

A graphical multi-response optimization technique was applied to determine the optimum combination of defatted rice bran content in the formula, barrel temperature profile and feed moisture content for the production of ready-to-eat breakfast cereal using a twin-screw extruder. The main criteria for constraint optimization were desirable dominant characteristics of ready-to-eat breakfast cereal – good physical properties such as expansion ratio, bulk density, water solubility index (WSI) and hardness, with better functional properties such as higher dietary fiber, total phenolic compounds and antioxidant activity. WSI is an important parameter for defining the application of extrudate as ingredients and in predicting how the material might behave if further processed especially in cereal product (Hashimoto and Grossmann, 2003). High WSI is related to higher levels of dextrinization encouraging stickiness of extruded products which lead the finished cereal product become slick on the surface and limp much more quickly during immersing in milk. These products are considered undesirable and inferior by consumers, an important fact which should be kept in mind during the production of ready-to-eat breakfast cereal. According to low WSI as required and other physical properties of the acceptable commercial ready-to-eat breakfast cereal products with similar cereal's shape as in this research, the following limits were proposed as follows: expansion ratio, 2.25-2.5; bulk density, 0.08–0.11 g/cm³; WSI, 26-30 and hardness, 1400-1700 g. In addition, the amount of total dietary fiber, 7.5-10%; total phenolic compounds, 125-145 mg GAE/100 g and antioxidant activity, 6.5-9.0 mmol Trolox/100 g were also proposed in order to use the higher possible range in this experiment. Superimposing the individual contour plots for the product response variables resulted in the identification of the overlaid area that satisfied all constraints. The grey portion of the overlaid area indicated the ranges of variables which could be considered to be the optimum ranges for desirable or best product quality as required in Figure 8.

Verification

Verification was carried out to confirm the suitability of the model equations for predicting optimum response values. The experimental results obtained under the selected conditions from RSM optimization and the predicted values using the model equations are shown in Table 3. The real experimental values were not statistically different

from the predicted values at a 95% confidence level. The results demonstrated the validation of the RSM model, indicating that the model was adequate for predicting optimum conditions in ready-to-eat breakfast cereal production enhanced with defatted rice bran.

Conclusion

Incorporation of defatted rice bran in extruded breakfast cereal not only utilized by-product from rice milling but also increased the value-added of commercialized product with health benefits. In this research, it is evident that the application of quadratic response surface methodology can be a practical and useful tool for optimization of the extrusion operating conditions of ready-to-eat breakfast cereal enhanced with defatted rice bran. Mathematical models from regression analysis can be used to interpret the relationship between the effect of extrusion operating variables on the physical and functional properties of the product. Good agreement between the values predicted and those determined experimentally confirm the adequacy of these models. Such information could help food processors predict the optimum manufacturing conditions and the performance of extruded materials to be used in novel products being developed in response to various consumer demands. Further work should be conducted deeply to characterize or purified with more concentrated source of functional component from defatted rice bran for enhanced more nutritional benefits and the promotion of healthy foods in extruded breakfast cereal production.

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