



Protein rich extruded products prepared from soy protein isolate-corn flour blends

Liang Yu^a, Hosahalli S. Ramaswamy^{a,*}, Joyce Boye^{a,b}

^a Department of Food Science and Agricultural Chemistry, McGill University, Macdonald Campus, 21111 Lakeshore, Ste Anne-de-Bellevue, PQ, H9X 3V9, Canada

^b Agriculture and Agri-Food Canada, St Hyacinthe, PQ, Canada

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ABSTRACT

Protein rich extruded products were prepared from soy protein isolate and corn flour blends using a twin screw extruder and the physical properties of the extruded product were evaluated and related to process variables: soy protein isolate (SPI) (32.2–66.6 g protein/100 dry matter), feed moisture (31.6–48.4 g/100 g) and process temperature (126.4–193.6 °C). A central composite rotatable design (CCRD) and response surface methodology was used to evaluate the significance of independent and interaction effects of extrusion process variables on the product's various physical properties (breaking stress, bulk density, expansion ratio, water solubility index, rehydration rate and color). Second order polynomial regression equations were developed to relate the product responses to process variables as well as to obtain the response surfaces plots. The independent variables had significant ($p \leq 0.05$) effects on physical properties of extrudates: (i) higher SPI and feed moisture contents increased the breaking stress and bulk density, but decreased the expansion ratio, water solubility index, and rehydration rate, (ii) higher SPI content decreased the color *L* value, whereas higher feed moisture content increased it, (iii) higher temperatures increased breaking stress, expansion ratio, rehydration rate and *L* value, but decreased the bulk density and water solubility index.

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1. Introduction

Since the consumer always demands healthy and nutritious foods, finding nutritional products is a food producer's unchangeable target. Degradation in the nutritional quality of finished food products arising from thermal processing of foods is a challenging matter under most traditional cooking methods. Extrusion cooking is a high temperature short time (HTST) cooking technique which provides thermal and shear energy to a food material to induce desirable physical and chemical changes. HTST extrusion process can minimize heat degradation of food nutrients, while improving digestibility by gelatinizing starch and denaturing protein (Harper, 1981) as compared to traditionally thermal processed foods. The extrusion cooking technique is preferable to others in terms of process continuity with high productivity and significant nutrient retention (Guy, 2001; Singh, Gamlath, & Wakeling, 2007). The functional properties of the food ingredients are modified during the extrusion processing process (Asp & Bjorck, 1989); extrusion

also destroys or inactivates the anti-nutritional or toxic compounds (i.e. trypsin inhibitors, hemagglutinins, and gossypol etc.), undesirable enzymes such as lipoxigenases, peroxidases, lipoxidases and lipases, microorganisms and other food-borne pests (Harper, 1981).

Extrusion of corn flour products has been extensively studied (Chinnaswamy & Hanna, 1988; Gomez & Aguilera, 1984); however, when such products are made exclusively from corn ingredient, they often lack in macro-nutrients like proteins. There has been a general interest in enriching the nutrient content of extruded foods by adding other nutritional ingredients to the feed mix used in the extrusion process (Guy, 2001; Konstance et al., 1998). Incorporating soy protein isolates (SPI) into corn flour can significantly increase its protein content and quality characteristics of the extruded product (Harper, Mercier, & Linko, 1989). With the development of the soybean processing industry, the soy crop has proven to be a low cost, and widely available source of superior quality protein. The use of SPI in extrusion products provides a high quality protein source, rich in lysine and bland in flavor, while reducing the flatulence factors and reducing sugars associated with whole soy flour. Thus SPI, widely used as a functional ingredient in the food industry, is believed to contribute to the overall improvement in extrusion product quality (Konstance et al., 1998).

* Corresponding author. Tel.: +1 514 398 7919; fax: +1 514 398 7977.
E-mail address: hosahalli.ramaswamy@mcgill.ca (H.S. Ramaswamy).

Showing a high potential to improve the nutritional profile of starch-based extruded food products (Sun & Muthukumarappan, 2002), SPI also has been credited with several potential health benefits. People with high (vs. low) soybean intakes have lower rates of coronary heart disease, breast cancer and osteoporosis (Liu, 2004; Sun & Muthukumarappan, 2002). Indeed, a statement that “including 25 g of soy protein per day in a diet low in saturated fat and cholesterol may reduce the risk of heart disease by lowering blood cholesterol levels” was approved by the US Food and Drug Administration (FDA, 1999). To uphold this soy protein health claim a single serving of the food must contain a minimum of 6.25 g of soy protein (FDA, 1999). Meeting the FDA soy protein threshold necessary to make such health claims can be achieved by producing SPI-enriched food products (i.e. extruded soy-corn blend food products) for all three meals and for snacks.

A number of studies have attempted to meet these goals. Konstance et al. (1998) produced extrudates combining corn meal with soy flakes, soy protein concentrate and soy oil. Faller, Klein, and Faller (1999) developed acceptable extruded snack products containing soy protein, and evaluated the influence of soy protein type, soy content, and moisture content. Muhungu et al. (1999) extruded corn flour and soy protein to investigate the influence of barrel temperature, moisture content, and relative residence time on extruder response and isoflavone profile. Sun and Muthukumarappan (2002) examined the effects of defatted soy flour content, feed moisture, screw speed and temperature on the functionality of soy-based extrudates. Seker (2005) evaluated the expansion ratio, bulk density and water solubility index of extrudates of SPI-modified corn starch mixtures. The effects of moisture content, screw speed and soybean content on the textural qualities of soybean-corn starch extrudates were studied by Li, Zhang, Jin, and Hsieh (2005).

In our previous study (Yu, Ramaswamy, & Boye, 2009), the effects of feed moisture, screw speed and barrel temperature on physical properties of extruded corn flour-SPI (20 g protein/100, dry matter basis) blends were studied to understand the influence of extrusion process on the product and to identify working range of parameters. Within the framework of this study, processing conditions (e.g., extrusion temperature, feed moisture and screw speed) were analyzed for constraint based optimization to provide end-products with a wide range of physical quality parameters (e.g., expansion ratio, bulk density, breaking stress, water solubility index, rehydration ratio and color). While the study showed very promising results producing high value products, the protein content was limited to a maximum of about 25 g/100 g (dry matter basis). This was because of the experimental design and somewhat broader objectives.

The previous experimental design was refined to include much higher levels of protein content (32.2–66.6 g/100 g dry matter basis) so that the product could be used as a protein supplement. In order to limit the experimental variables to 3, only two of the three factors from the previous study (moisture content and temperature) were employed in this study so that the same 20 run CCRD design could be employed. While the study may look similar, the influencing parameters and their ranges selected were quite different (temperature in the range of 126.4–193.6 °C and feed moisture content in the range of 31.6–48.4 g/100 g (wet basis) as compared with the earlier 140–180 °C, and 18–38 g/100 g, respectively) making the study and the product significantly different. The overall objectives were to evaluate the effects of processing parameters (protein content, feed moisture content, and extrusion processing temperature) on the quality of extruded protein rich products and assess their optimization scenarios.

2. Materials and methods

2.1. Materials

Corn flour from Brar Natural Flour Mills (Winnipeg, MB) was purchased from a local market. The composition of the flour was: fat, 1.7 g/100 g (wet basis); carbohydrate, 76.7 g/100 g (wet basis); protein, 10 g/100 g (wet basis) and moisture, 12 g/100 g (wet basis). Soy protein isolate containing protein, 90 g/100 g (wet basis); carbohydrates, 5 g/100 g (wet basis) and moisture 5 g/100 g (wet basis) was obtained from American Health & Nutrition (Ann Arbor, MI), while soy flour containing protein 40 g/100 g (wet basis); fat, 22 g/100 g (wet basis); carbohydrates 33.5 g/100 g (wet basis) and moisture, 4.5 g/100 g (wet basis) was obtained from Soyador (Quebec, Canada). The purpose of adding soy flour to the mixture was to provide some natural fat for lubrication of the extruder during the process. To a small extent some fat also came through corn flour. The moisture contents of all the flours were measured before mixing. The soy flour used was full fat regular ground flour. The amount of soy flour added was relatively small to give a calculated 2 g/100 g (dry basis) of fat in the product. It was found necessary in our laboratory system to have this fat in the feed mix so that the extrudates emerge out easily. We assumed it was due to the lubrication and resulting in less stickiness problem during clean-up.

2.2. Extrusion process

A co-rotating twin screw extruder (DS32-II, Jinan Saixin Food Machinery, Shandong, P. R. China) was used in all extrusion processes. The barrel was equipped with four independent temperature controlled zones. The first zone (after the feeding section) temperature was controlled at 110 °C, second and third zones (mixing part) were controlled at 135 °C and 150 °C, the temperature of the fourth barrel zone (metering section) was adjusted to the required levels as one of the variables. The diameter of the screw was 30 mm. The length to diameter ratio of the extruder barrel was 20:1. The diameter of the hole in the die was 5 mm with a die length of 27 mm. A constant screw speed of 100 revolutions per minute (rpm) was selected based on previous experimental results, and to limit the number of process variables to three. The extruder was fed automatically through a conical hopper, keeping the flights of the screw fully filled and avoiding accumulation of the material in the hopper.

After stable conditions were established, extrudates were collected and cut into 35 mm long cylindrical specimens and air dried at 55 °C for 120 min by a convection oven with an air velocity of 0.1 m/s as measured by an anemometer to a moisture content of 9–10 g/100 g wet basis. Dried samples were stored in air-tight plastic containers at room temperature until analysis.

2.3. Experimental design of soy protein isolate and corn starch blend extrusion

In a previous experiment (Yu et al., 2009), different variables including screw speed, moisture content and barrel temperature were tested, in order to get a protein enriched product by maintaining a 20 g/100 g SPI level in the feed mixture. In this study, protein content was used as one of the prime variable and two other independent variables (barrel temperature and moisture content) were selected and investigated using a central composite rotatable design (CCRD) (Draper, 1982). Protein content was varied from 32.2 to 66.6 g/100 g (dry basis); feed moisture content from 31.6 to 48.4 g/100 g (wet basis); and extrusion barrel temperature (metering section) from 126.4 to 193.6 °C. Overall, 20 experimental runs were made, each with 8 (2^3) factorial points (three level for

each variables), six star corner points (two for each variable) and 6 center points to meet the statistical design requirements. The CCRD experiment ranges for the 3 independent variables were selected based on preliminary tests.

An Excel worksheet was used to determine the quantities of the SPI, corn flour and moisture (based on a mass balance approach). The details of the different test run with coded (and real) values of the process variables as well as the amount of ingredients added for a 2.0 kg batch (excluding moisture) are shown in Table 1. First step was to fix the total feed mix to be 2.0 kg (excluding the water to be added), then fix wet basis moisture content at the desired level. This will enable computation of the quantity of water to be added (first estimate). Then the oil and protein contents were set at the required level, then the quantity of SPI and soy flour to be used (balance is corn flour to make 2 kg batch size) are sequentially increased until the desired dry basis levels of protein and oil are converged to the target levels. The calculated data are summarized in Table 1. It is relatively easy to do this on an excel spread sheet.

The flours were mixed in a Hobart mixer (Hobart Food Equipment Group Canada, North York, ON) operating at a medium speed. Predetermined amount of water was added to adjust the mixture to the desired moisture content, according to the experimental design. The blend was mixed for 20 min in the Hobart mixer before use.

In order to develop the RSM models, coded values (P_{cv} , T_{cv} , M_{cv} , respectively) were derived from the numerical values of the independent variables protein content (P , as g/100 g), barrel temperature (T , as °C), and feed moisture (M , as g/100 g):

$$P_{cv} = \frac{(P - 50) \times 1.682}{10} \quad (1)$$

$$T_{cv} = \frac{(T - 160) \times 1.682}{20} \quad (2)$$

$$M_{cv} = \frac{(M - 35) \times 1.682}{10} \quad (3)$$

2.4. Physical properties

2.4.1. Breaking stress

Breaking stress (BS) was measured in a 3-point bend test (Zasyplin & Lee, 1998) using the TA.XT Plus Texture Analyzer

(Texture Technologies Corp., Scarsdale, NY/Stable Micro Systems, Godalming, Surrey, UK.) equipped with a 50 N load cell. The extruded product was placed on two rounded stands (bridge) 30 mm apart. A rounded plunger was made to push the sample at the middle of the bridges at 5 mm/min until breakage occurred. BS was determined as the breaking force per unit cross section area (N/mm²). Eight measurements were made on each product (separate samples) and their mean value was used.

2.5. Bulk density

Bulk density (ρ , g/ml) was measured using the displacement method (Seker, 2005). Extrudates strands were cut into roughly 25 mm sections (≈ 15 g) and weighed (M_{ext} , g). Each strand was then placed in a graduated cylinder, to which a certain volume (V_{om} , ml) of yellow millet particles were added, and the final volume was measured (V_{ym} , ml). BD was calculated as:

$$\rho = \frac{M_{ext}}{V_{ym} - V_{om}} \quad (4)$$

2.5.1. Expansion ratio

Expansion ratio (ER) is defined as the ratio of the diameter of the extrudate to the diameter of the die (Jyothi, Sheriff, & Sanjeev, 2009). In order to determine the ER, 20 randomly selected segments of each sample were measured using vernier caliper and the mean value was used.

2.5.2. Water solubility index

The water solubility index (WSI) of the extrudate was determined following the method described by Anderson, Conway, Pfeife, and Griffin (1969) with little modification. The extrudates were ground and about 2 g (W_{os}) of the ground extrudate was placed into a centrifuge tube. Distilled water (25 ml) at 30 °C was added with the sample. After 10 min standing with intermittent shaking every 2 min, the sample was centrifuged at 4000× g for 15 min (Sorvall GLC-2B General Laboratory Centrifuge, Du Point Instrument). The supernatant was decanted into a Petri dish and dried at 105 °C overnight. The weight of the dry solid (W_{ds}) was determined and the WSI (g/100 g) calculated:

$$WSI = \frac{W_{ds}}{W_{os}} \times 100 \quad (5)$$

2.5.3. Rehydration ratio

An air dried sample (≈ 20 g) of extrudate were weighed (M_1) and placed into 500 ml of water at 30 °C for 15 min. The water was then drained and the rehydrated sample was weighed (M_2). The rehydration ratio (RR, g/100 g) was defined as:

$$RR = \frac{M_2 - M_1}{M_1} \times 100 \quad (6)$$

2.5.4. Color

Color measurements were done using a Minolta colorimeter CM-500d using an aperture of 1.2 cm diameter. Samples were ground and passed through a standard sieve (#50) prior to the color determination. The instrument was calibrated with a standard white tile ($L = 77.58$, $a = -0.27$, $b = -26.63$). The color was reported in terms of Hunter L , a and b values. Eight measurements on each sample were taken and the mean value was used.

Table 1

Experimental (CCRD) design with coded and (actual) values for soy protein (P), moisture content (M), and processing temperature (T), along details of the quantities of soy protein isolate (SPI), soy flour, corn flour and water added (2.0 kg batch).

Run	P (g/100 g db)	M(g/100 g wb)	T (°C)	SPI (g)	Soy flour (g)	Corn flour (g)	Water (g)
1	-1 (40)	-1 (35)	-1 (140)	633	67	1300	790
2	1 (60)	-1 (35)	-1 (140)	1095	108	797	842
3	-1 (40)	1 (45)	-1 (140)	633	67	1300	1298
4	1 (60)	1 (45)	-1 (140)	1095	108	797	1359
5	-1 (40)	-1 (35)	1 (180)	633	67	1300	790
6	1 (60)	-1 (35)	1 (180)	1095	108	797	842
7	-1 (40)	1 (45)	1 (180)	633	67	1300	1298
8	1 (60)	1 (45)	1 (180)	1095	108	797	1359
9	-1.68 (33.2)	0 (40)	0 (160)	478	53	1469	1004
10	1.68 (66.6)	0 (40)	0 (160)	1252	122	626	1098
11	0 (50)	-1.68 (31.6)	0 (160)	862	87	1051	676
12	0 (50)	1.68 (48.4)	0 (160)	862	87	1051	1547
13	0 (50)	0 (40)	-1.68 (126.4)	862	87	1051	1051
14	0 (50)	0 (40)	1.68 (193.6)	862	87	1051	1051
15	0 (50)	0 (40)	0 (160)	862	87	1051	1051
16	0 (50)	0 (40)	0 (160)	862	87	1051	1051
17	0 (50)	0 (40)	0 (160)	862	87	1051	1051
18	0 (50)	0 (40)	0 (160)	862	87	1051	1051
19	0 (50)	0 (40)	0 (160)	862	87	1051	1051
20	0 (50)	0 (40)	0 (160)	862	87	1051	1051

2.6. Regression modeling and statistical analysis

The second order polynomial equation fitted with coded variables was:

$$Y = B_0 + \sum_{i=1}^n B_i X_i + \sum_{i=1}^n B_{ii} X_i^2 + \sum_{i=1}^{n-1} \sum_{j=i+1}^n B_{ij} X_i X_j \quad (7)$$

where, Y represents the experimental responses (BS, BD, ER, WSI, RR and color), B_0 , B_i , B_{ii} and B_{ij} are constants and regression coefficients of the model, while X_i and X_j are independent extrusion processing variables, n is number of processing variables (here $n = 3$) The whole model includes linear, quadratic and cross-product terms.

Experimental data were analyzed using Design-Expert 6.0 (Stat-Ease Inc. Minneapolis, MN) and Microsoft Excel 2003 Version. Design-Expert was used to solve the second order polynomial regression equation and Excel was used to investigate the effects of three independent input variables of SPI content, feed moisture content and extrusion processing temperature on the physical properties (BS, BD, ER, WSI, RR and color) of extrudates. Analysis of variance (ANOVA) was employed to justify the model fitness as well as significant effects of independent variables on responses.

3. Results and discussion

The experiment arrangements (test run number) and data on the physical properties of breaking stress (BS), bulk density (BD), Expansion ratio (ER), water solubility index (WSI), rehydration rate (RR) and color (L , a , b values) of extruded products are shown in Table 2. The table also indicates the approximate proximate composition (on dry weight basis) of the product obtained after the extrusion process. Second order polynomial regression equations were established by considering significant factors on the basis of t (t -test) > 2.5 at probability level $p < 0.05$ and coefficients obtained are presented in Table 3. The established equations show empirical relationship between physical properties (BS, BD, ER, WSI, RR and Color) and the coded value of independent variables of total protein content (P), feed moisture content (M) and barrel temperature (T). An analysis of variance (ANOVA) was done to analyze the impacts of independent variables of soy protein isolate

content, feed moisture content and temperature on the physical properties of BS, BD, ER, WSI, RR and the color parameter L of extruded product.

The ANOVA study results showed that total protein content significantly affected the WSI, ER and L ($p < 0.05$). Feed moisture content (M) significantly ($p < 0.05$) affected the RR, BD and BS. Barrel temperature (T) significantly ($p < 0.05$) affected WSI, RR, BD, BS and ER. All the three parameters showed the quadratic effect ($p < 0.05$) to WSI, BS, BD and ER. Interactive effects of total protein content and feed moisture were found on RR, BS, BD and L ($p < 0.05$). Interactive effects ($p < 0.05$) of total protein content and barrel temperature were found on RR, BS, BD and ER. Interactive effects ($p < 0.05$) of moisture content and barrel temperature were found on WSI, RR, BS, BD and ER.

Various physical properties have been studied in different extruded products. Jyothi et al. (2009) studied the physical properties including bulk density, true density, porosity, and expansion ratio; water absorption index, water solubility index, oil absorption index, in the single extruder to process tuber starch. Rocha-Guzman et al. (2008) studied water absorption index (WAI), water absorption capacity (WAC), oil absorption capacity (OAC), and emulsifying capacity (EC) in the extrusion process of bean cultivars flour. Özer, İbanoğlu, Ainsworth, and Yağmur (2004) studied the physical properties (bulk density, expansion, and porosity) of a nutritionally balanced extruded snack food by the RSM method. The influence of process variables on physical properties have been shown to be generally significant in all these studies.

Table 2 also provides some data on the resulting product in the form of proximate composition and the actual water content of the products. These are predicted values based on the dry ingredients and the moisture content of the product as it exited from the extruder. It can be seen that the extruded product has a protein content in the range 40–60 g/100 g (dry basis); carbohydrate in range 31–65 g/100 g (dry basis) and moisture content in the range 22–35 g/100 g (wet basis). While they provide a protein and carbohydrate rich product, the extruded product is also too high in moisture content to provide adequate stability. The water activity in most cases was higher than 0.85 and hence the drying of extruded product for few hours was necessary to produce a low moisture shelf stable product.

Table 2
Experimental results for each test run (details in Table 1) as means (and standard deviation) of physical properties of soy protein isolate- corn starch blend extrusions and approximate proximate composition (in dry matter) and the water content of the extruded product [BS = breaking stress, BD = bulk density, ER = expansion ratio, WSI = water solubility index, RR = rehydration ratio, L , a , b (color)].

Run	BS (N/mm ²)	BD (g/ml)	ER	WSI (g/100 g)	RR (g/100 g)	L	a	b	Protein (g/100 g, db)	Carbohydrate (g/100 g, db)	Moisture (g/100 g, wb)
1	0.641 (0.017)	0.771 (0.037)	1.31 (0.048)	3.6 (0.127)	58 (2.1)	87.01 (3.0)	0.12 (0.004)	23.85 (0.84)	40.1	57.9	30.7
2	0.246 (0.033)	0.864 (0.039)	1.43 (0.078)	4.7 (0.238)	195 (9.5)	75.17 (4.0)	1.54 (0.083)	28.77 (1.59)	60.0	38.0	29.0
3	0.179 (0.012)	0.528 (0.027)	1.35 (0.053)	5.5 (0.321)	49 (3.8)	83.75 (3.2)	1.79 (0.068)	31.52 (1.14)	40.1	57.9	32.0
4	0.141 (0.027)	0.723 (0.098)	1.45 (0.204)	6.8 (0.763)	95 (10.3)	78.47 (9.0)	3.39 (0.667)	32.68 (2.46)	60.0	38.0	34.4
5	0.523 (0.032)	0.613 (0.036)	1.37 (0.076)	4.9 (0.291)	157 (8.5)	84.89 (5.0)	0.18 (0.011)	25.11 (1.50)	40.1	57.9	23.6
6	0.458 (0.059)	0.423 (0.075)	1.69 (0.219)	5.1 (0.641)	141 (12.8)	74.21 (8.8)	3.47 (0.218)	35.36 (3.28)	60.0	38.0	24.0
7	0.332 (0.009)	0.656 (0.019)	1.28 (0.021)	3.1 (0.046)	205 (4.4)	82.87 (1.2)	1.57 (0.023)	32.15 (0.46)	40.1	57.9	26.7
8	0.773 (0.051)	0.514 (0.028)	1.51 (0.094)	3.8 (0.195)	96 (7.1)	83.03 (4.5)	1.39 (0.077)	24.79 (1.03)	60.0	38.0	27.5
9	0.431 (0.015)	0.662 (0.016)	1.41 (0.046)	2.9 (0.101)	132 (7.2)	87.37 (3.0)	1.05 (0.037)	34.72 (1.21)	33.2	64.8	30.5
10	0.282 (0.021)	0.641 (0.055)	1.59 (0.107)	4.6 (0.308)	122 (5.5)	75.69 (5.1)	2.97 (0.219)	29.29 (1.95)	66.6	31.4	29.2
11	0.586 (0.028)	0.669 (0.034)	1.45 (0.054)	3.8 (0.138)	131 (5.6)	84.24 (3.3)	0.52 (0.019)	25.17 (0.91)	50.0	48.0	24.2
12	0.216 (0.007)	0.541 (0.018)	1.49 (0.033)	4.5 (0.108)	96 (2.7)	84.38 (2.3)	0.55 (0.013)	23.09 (0.56)	50.0	48.0	33.7
13	0.237 (0.010)	0.791 (0.035)	1.25 (0.066)	5.6 (0.202)	60 (2.7)	84.38 (3.2)	0.79 (0.035)	23.73 (1.03)	50.0	48.0	31.8
14	0.726 (0.020)	0.452 (0.023)	1.51 (0.041)	3.9 (0.109)	195 (5.2)	80.13 (2.2)	1.52 (0.043)	29.07 (0.82)	50.0	48.0	23.8
15	0.684 (0.031)	0.475 (0.021)	1.74 (0.067)	3.4 (0.182)	107 (5.5)	81.81 (4.3)	1.43 (0.077)	25.34 (1.99)	50.0	48.0	26.8
16	0.816 (0.038)	0.478 (0.019)	1.73 (0.061)	2.9 (0.127)	129 (5.9)	80.13 (3.2)	1.86 (0.075)	28.97 (1.18)	50.0	48.0	30.3
17	0.671 (0.062)	0.466 (0.058)	1.77 (0.098)	3.4 (0.221)	139 (10.3)	80.03 (5.2)	0.34 (0.022)	24.15 (1.54)	50.0	48.0	28.5
18	0.721 (0.053)	0.495 (0.036)	1.75 (0.105)	2.7 (0.163)	139 (10.7)	80.19 (4.8)	0.55 (0.043)	24.79 (1.49)	50.0	48.0	27.9
19	0.828 (0.041)	0.476 (0.028)	1.69 (0.093)	3.1 (0.171)	139 (7.4)	79.68 (4.4)	0.68 (0.057)	25.85 (1.42)	50.0	48.0	28.2
20	0.818 (0.034)	0.479 (0.101)	1.76 (0.114)	3.3 (0.214)	129 (8.4)	81.21 (5.2)	1.05 (0.046)	25.65 (1.66)	50.0	48.0	28.0

Table 3

Regression equations for physical properties of soy protein isolate-corn starch blend extrusion (taking significant parameters on the basis of $t > 2.5$ at probability level $p \leq 0.05$). [BS = breaking stress, BD = bulk density, ER = expansion ratio, WSI = water solubility index, RR = rehydration ratio, L = lightness (L , a , b color space)].

Physical properties	Equations (in coded values)
BS (N/mm ²)	$Y_{BS} = 0.756 - 0.078P + 0.125T - 0.137P^2 - 0.122M^2 - 0.093T^2 + 0.108P * M + 0.101P * T + 0.086M * T$
BD (g/ml)	$Y_{BD} = 0.478 - 0.034M - 0.092T + 0.062P^2 + 0.045M^2 + 0.051T^2 + 0.019P * M - 0.078P * T + 0.065M * T$
ER	$Y_{ER} = 1.74 + 0.079P + 0.055T - 0.087P^2 - 0.097M^2 - 0.129T^2 + 0.041P * T - 0.041M * T$
WSI (g/100 g)	$Y_{WSI} = 0.031 + 0.005P - 0.05T + 0.003P^2 + 0.004M^2 + 0.007T^2 - 0.009M * T$
RR (g/100 g)	$Y_{RR} = 125.7 - 12.1M + 31.4T - 23P * T - 38.5P * T + 14.1M * T$
L value (color)	$Y_L = 81.332 - 3.462P + 2.175P * M$

P: soy protein; M: moisture content; T: barrel temperature.

3.1. Breaking stress (BS)

The values of breaking stress (BS) of extruded products under experimental conditions are presented in Table 2. The highest value

of BS was 0.828 N/mm² while extrusion was done at 160 °C with 50 g/100 g (dry basis) protein content and 40 g/100 g (wet basis) feed moisture. The lowest value of BS was 0.141 N/mm² while extrusion processing at 140 °C with 40 g/100 g (dry basis) protein content and 45 g/100 g (wet basis) feed moisture. According to the BS, barrel temperature is the most significant affecting parameter ($p < 0.05$). Response surface plot for protein content vs. moisture for temperatures of 140 °C, 160 °C and 180 °C as predicted by the model are shown in Fig. 1. The results show that with an increase in temperature, the BS values increased, especially for the high protein and high moisture content product. The lowest BS appeared when the barrel temperature was the lowest (Fig. 1A), the moisture and protein content are at the highest level; the highest BS appeared in Fig. 1C, when the barrel temperature is the highest, the moisture and protein content are also at the highest level.

Barrel temperature and material moisture content significantly ($p \leq 0.05$) affected the BS and all the quadratic effects including protein content, moisture content and barrel temperature, and interaction effects including protein content and moisture content, protein content and temperature content, moisture content and temperature significantly affected BS. Lack of fit was not significant relative to the pure error, which meant the model was well fitted. The regression equation for the empirical relationship between BS

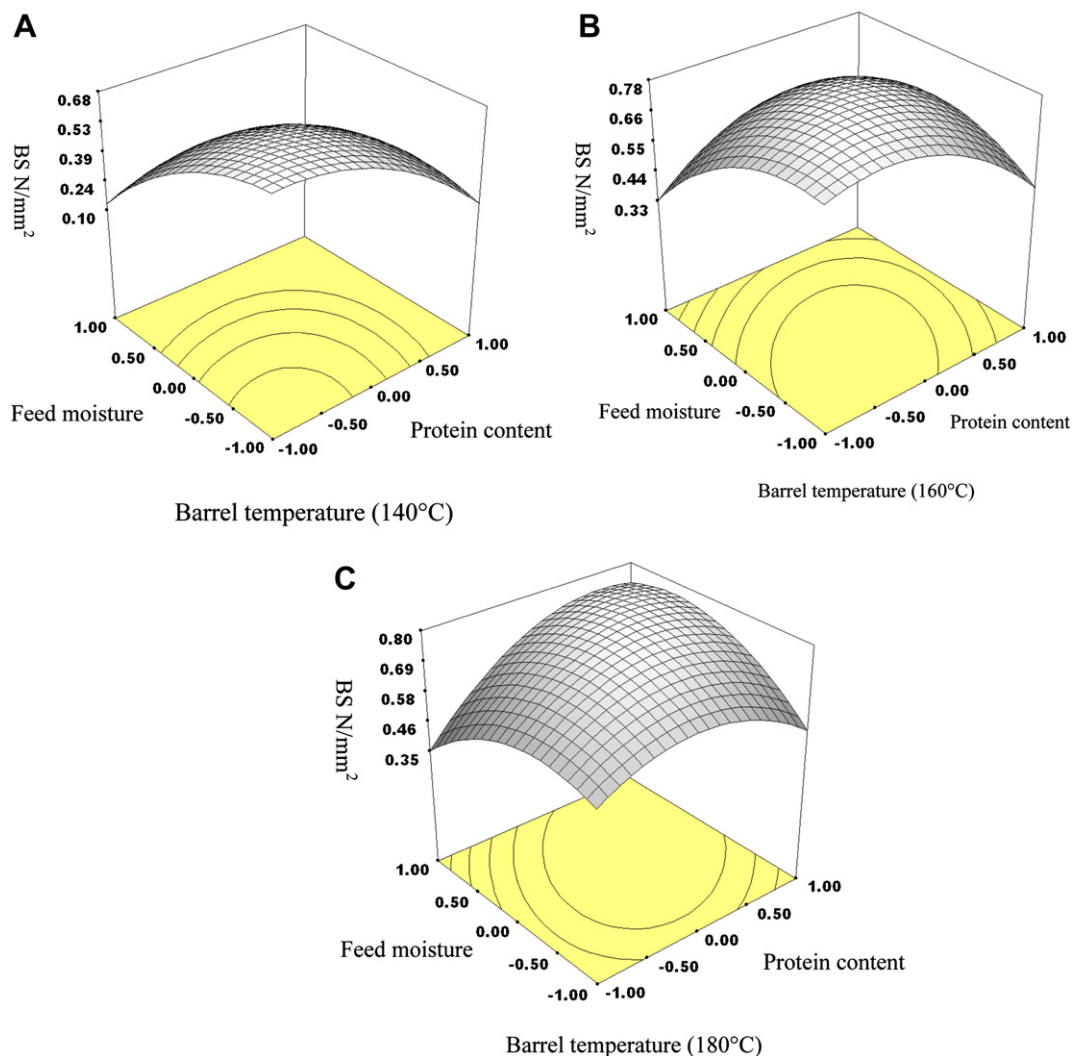


Fig. 1. Response surface plot of breaking stress (BS) of soy protein isolate-corn starch blend extrusion for the effect of soy protein content and moisture content at different barrel temperatures.

(Y_{BS}) and the independent extrusion processing variables in coded form is shown in Table 3.

Sun and Muthukumarappan (2002) revealed that the shear force per unit weight of extrudate decreased with an increase in the feed moisture and a decrease in barrel temperature. It may be expected that high moisture content in the blends with high temperature extrusion processing expands the products due to release of superheated steam. This phenomenon helps to make hollowed and low density products that decrease the breaking stress of the extruded products. But our study revealed the reverse findings regarding to high moisture, high protein content and high barrel temperature of the process. One reason can be the high density product naturally offers high breaking stress. A high extrusion processing temperature with a high screw speed provides a high level of thermal and mechanical energy simultaneously, which possibly leads to excessive structural damage and breakdown, and hence density increases slightly (Guha, Zakiuddin Ali, & Bhattacharya, 1997). Another reason can be the air cell membrane of the extrudates became harder due to high soy protein isolate content. Harper (1981) showed that shear strength of the extrudates increased with an increase in the protein content and processing temperature. ANOVA study showed that, between the three parameters, barrel temperature was the most significant one affecting the BS, and with a combination effect of the three parameters, the result was even more intense.

3.2. Bulk density (BD)

The experimental values of bulk density of extrudates under different designed extrusion conditions are presented in Table 2. BD ranged from 0.423 to 0.864 g/ml for the extruded products. The response surface plots were prepared (not shown to reduce the manuscript size; instead the one for ER which causes the bulk density to be lower is presented next) to evaluate and visualize the effect of feed moisture and protein content effect on BD under different temperatures. Results showed that at the low barrel temperature (140 °C), increasing protein content and decreasing moisture content resulted in a higher BD. At high barrel temperature (180 °C), increasing protein content lead to decrease the BD. At middle barrel temperature (160 °C), the minimum BD area appeared in the middle part of the figure.

Soy protein isolate (SPI) constructs small uniform pores in the extruded products after being squeezed out of the die as soy protein isolate can work as high quality emulsifiers between hydrophilic materials and hydrophobic materials by exposing the hydrophilic groups and hydrophobic groups to their respective phases (Li et al., 2005). The thickness of the wall of the pores become thinner when the amounts of soy protein isolate increase and the soy protein isolate absorbs high amounts of water. Thus, it is logical to expect an increasing bulk density of the extrudates with increasing protein and feed moisture contents. Starch gelatinization during extrusion processing has a big influence on bulk density of extrudates. The low

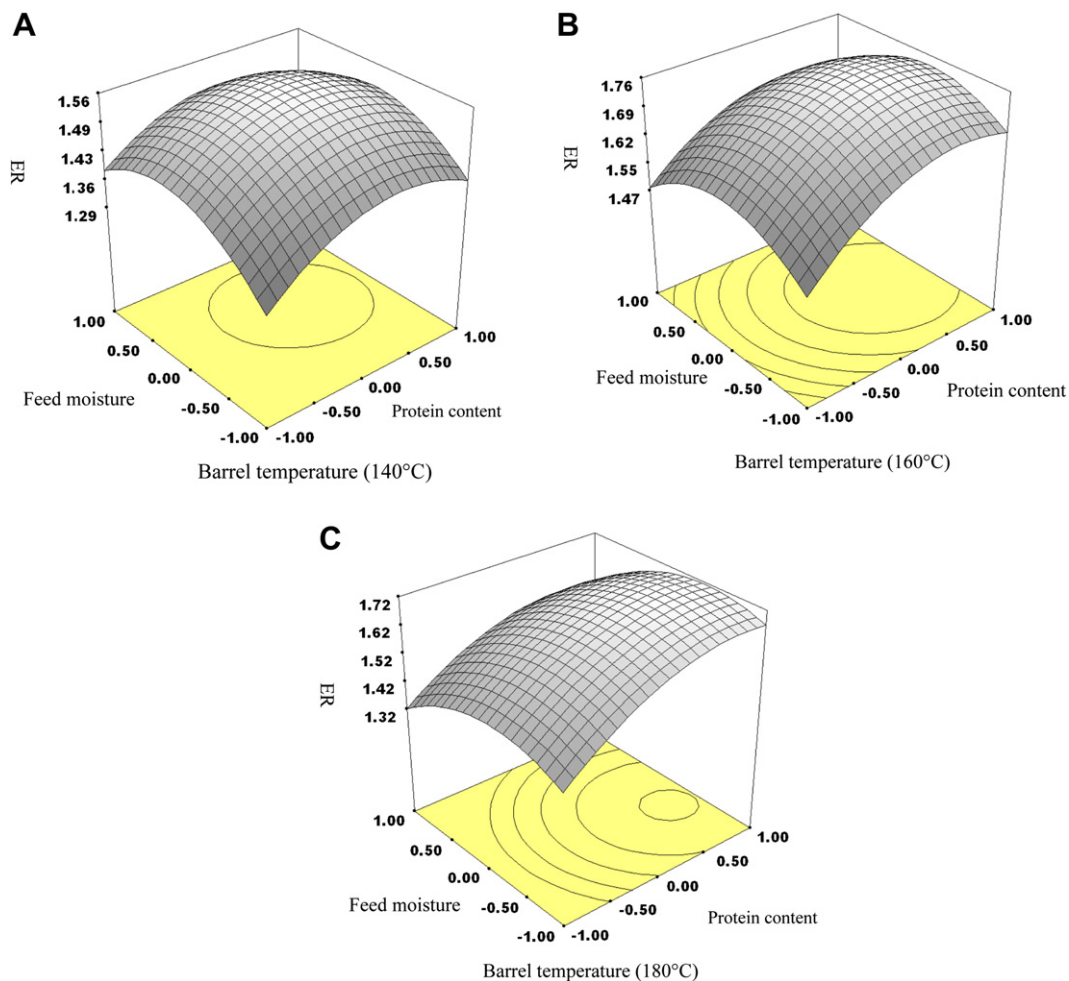


Fig. 2. Response surface plot of expansion ratio (ER) of soy protein isolate-corn starch blend extrusion for the effect of soy protein content and moisture content.

processing temperature decreases the extent of gelatinization of the extruded products, which leads to low swelling and low volume of extrudates, therefore, bulk density of extrudates increases with the decreasing of extrusion processing temperature.

The regression equation for bulk density (Y_{DB}) is also shown in Table 3. ANOVA analysis shows that both barrel temperature and moisture content are significantly affect BD in linear and quadratic forms. Protein content significantly affected BD through a quadratic model. All the interaction effect among protein content, barrel temperature and moisture content were significant for BD. This explains that temperature provides a curvilinear effect on extrudate bulk density and its quadratic effect dominates at high temperature (Guha et al., 1997).

3.3. Expansion ratio (ER)

Expansion ratio (ER) indicates the extent of puffing of extruded products. The experimental values of expansion ratio of extrudates under different designed extrusion conditions are shown in Table 2. The values of expansion ratio (ER) varied from 1.25 to 1.77 on the basis of combinations of extrusion process variables. The highest expansion ratio was 1.77 for extrusion processing at 160°C with 50 g/100 g protein content and 40 g/100 g feed moisture content; on the other hand, the lowest expansion ratio was 1.25 for extrusion processing at 126.4°C with the same protein and feed moisture contents.

The response surface plot presented [Fig. 2 (A:140 °C, B:160 °C, C:180 °C)] showed that the expansion ratio of extrudates increased

with an increasing of feed moisture content and after reaching a maximum the ER decreased with further increasing in moisture content. Similar trends were also observed with protein content and barrel temperature.

The regression equation for the relationship between expansion ratio (Y_{ER}) and independent variables in terms of coded variables is showed in Table 3. The analysis of variance showed that all the quadratic effects from protein content, feed moisture content and barrel temperature were significant with ER. The interaction effect between protein content and barrel temperature, moisture content and barrel temperature were significant with ER. Protein content was the most significant factor affecting ER. Moisture content was less significant than temperature. This result is some different with our prior study (Yu et al., 2009) in which feed moisture content showed more significant effect to ER than barrel temperature. The difference could be because the protein content was lower and maintained below 25 g/100 g (dry basis). In this study, protein content is much higher, and the combination of protein and moisture content became highly related with the process temperature and the protein content, so the barrel temperature became more significant.

3.4. Water solubility index (WSI)

The results of water solubility index (WSI) for different experimental conditions are showed in Table 2. The water solubility index (WSI) of extrudates ranged between 2.7 g/100 g (protein content

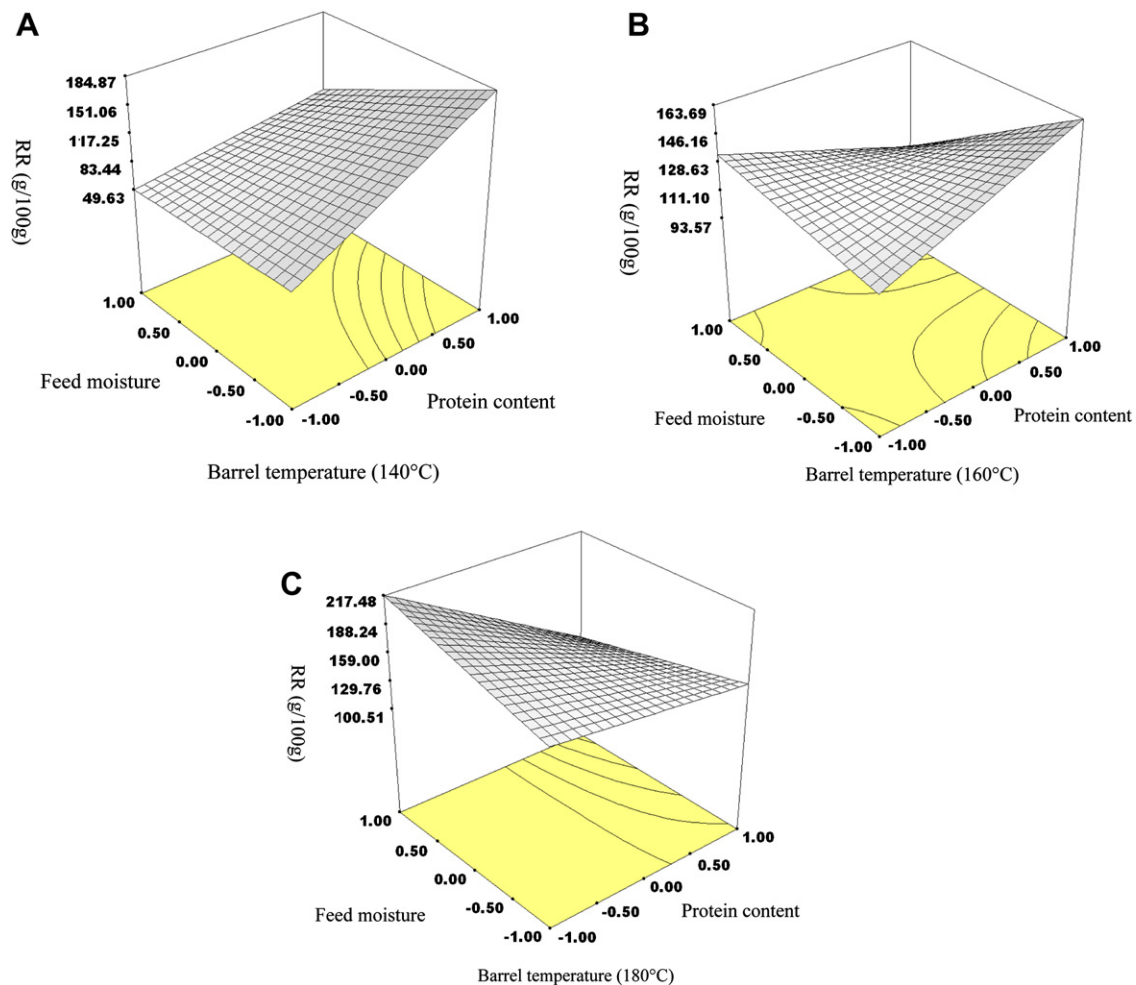


Fig. 3. Response surface plot of rehydration ratio (RR) of soy protein isolate-corn starch blend extrusion for the effect of soy protein content and moisture content.

50 g/100 g, moisture content 40 g/100 g (wb), barrel temperature 160 °C) and 6.8 g/100 g (protein content 60 g/100 g, moisture content 45 g/100 g (wb), barrel temperature 140 °C).

The response surface plot of water solubility index were likewise prepared but not shown. Under lower barrel temperature (140 °C), the WSI continued to decrease as the feed moisture and protein content decreased. With higher barrel temperature (180 °C) the WSI increased with a decrease in the feed moisture and with an increase in the protein content. When the barrel temperature is in the middle (160 °C), WSI showed the lowest value in the middle part.

ANOVA analysis demonstrated that the model was significant ($p < 0.05$). Again barrel temperature and protein content significantly affected the WSI through a linear model. All three factors were significant for WSI with quadratic model, and the interaction between barrel temperature and moisture content was also significant ($p < 0.05$). The regression equation, showing empirical relationship between water solubility index (Y_{WSI}) and the processing variables are shown in Table 2.

In our previous study (Yu et al., 2009), moisture content was the most significant factor that affected the WSI, and a higher moisture content resulted in a higher WSI. The reason for this might be at lower feed moisture levels, it is possible that there was not enough water for the starch gelatinization and protein denaturation to be completed. This could be the reason for gradual increase in WSI with an increase in moisture content. In

this study, when the barrel temperature is at the lower level (140 °C), the trend between moisture content and WSI is the same as the previous study, but when the barrel temperature was increased to a higher level (180 °C), the WSI decreased with the increasing of the moisture content. Hagenimana, Ding, and Fang (2006) reported that with extruded rice flour when feed moisture increased from 16 to 22 g/100 g (wet basis), WSI decreased. Gomez and Aguilera (1984) concluded that low feed moisture content of extrudate reduced starch gelatinization and shear degradation of starch, which reduced the physical breakdown of the granules. This may be a possible reason why water solubility index of extrudates increased with the decreasing of feed moisture content. Cumming, Stanley, and Deman (1973) concluded that the high temperature in the extrusion processing caused most of the water soluble protein to break into small subunits, or become insoluble, and/or be redistributed. This phenomenon may cause to decrease the water solubility index with increasing temperature and soy protein isolate. Different results indicate that the key factor that affects the WSI is how much of the water soluble protein turns into water insoluble. In this study it has been shown that, at the lower temperature condition, increasing water content will increase the starch gelatinization and the protein denaturation, but when the temperature is higher to 180 °C, the water soluble protein begin to break into insoluble subunits hence the adding water content resulted the decrease of the WSI.

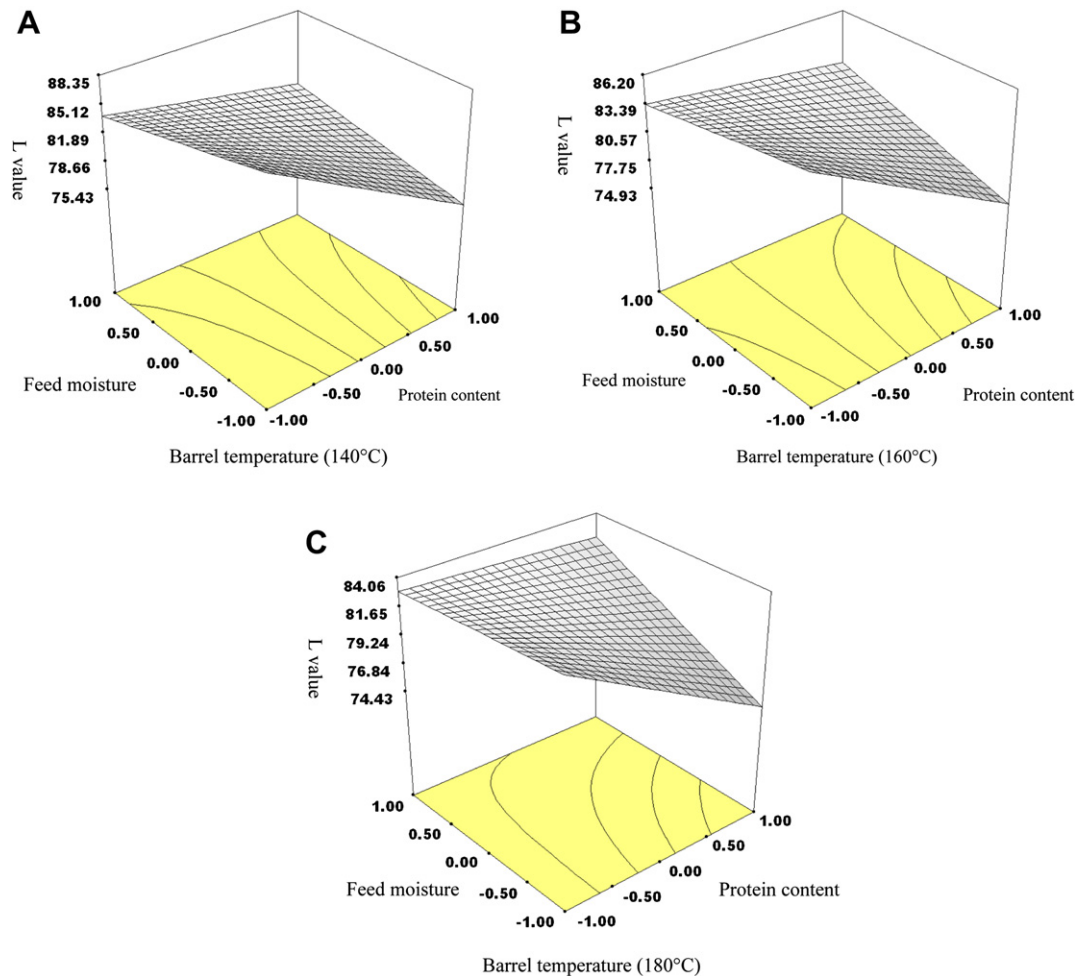


Fig. 4. Response surface plot of color (L value) of soy protein isolate-corn starch blend extrusion for the effect of soy protein content and moisture content.

3.5. Rehydration ratio (RR)

Rehydration ratio of extrudates extruded for different experimental conditions are listed in Table 2. The values of RR varied from 49 g/100 g to 205 g/100 g. The regression equation for the relationship between rehydration ratio (Y_{RR}) and independent variables is shown in Table 3. Barrel temperature and material moisture content were significant for RR, temperature was the most significant factor and all interaction among protein content, material moisture content and barrel temperature were also significant.

The response surface plot of rehydration ratio of extrudates are presented in Fig. 3 (A:140 °C, B:160 °C, C:180 °C). These figures showed RR have more complicated trend under different extrusion conditions. Under lower barrel temperature (140 °C), higher protein content resulted in higher RR. When the temperature was increased to 160 °C, an increase protein content at high moisture content (45 g/100 g, dry basis) slightly decreased the RR, but increase protein content at a up to a high low moisture content (35 g/100 g, dry basis) increased the RR. With barrel temperature at 180 °C, RR decreased with an increase in protein content.

Harper (1981) reported that low moisture content in feed decreased the trypsin inhibitor and increased rehydration rate of extrudates. Increasing feed moisture content may lead to retain high water content inside the extruded products and consequently decrease the rehydration ratio. The high processing temperature probably creates more open spaces and air cells in the product structure due to high temperature generates high thermal energy, which increases the level of superheated steam during extrusion processing. This may impart to imbibe more water when rehydrating the extrudates, subsequently, rehydration rate of extrudate increases with the increasing of extrusion processing temperature. Protein content in feed decreases the starch molecular degradation, an increases in protein content with relative decrease in starch content may influence the extent of starch gelatinization during extrusion processing leading to a decrease in water absorption relatively (Yagci & Gogus, 2008). This

phenomenon may cause the decrease of rehydration rate of extrudate with an increase in soy protein isolate content in feed blends.

3.6. Color

Color is one of the most vital attributes of any food product due to consumer acceptability considerations. The color of extruded products was measured in terms of Hunter L (lightness), a (redness), and b (yellowness) values. The values of L , a , and b under different designed experimental conditions are given in Table 2. The results indicated that the L values of extrudates varied between 74.2 and 87.4, a -values ranged from 0.12 to 3.47 and b values varied from 23.1 to 35.4. L value increased with decreasing soy protein isolate content and increasing processing temperature. The a -value increased with increasing protein content and process temperature. The highest b value was found while processing was done at 180 °C with 60 g/100 g (dry basis) soy protein isolate and 35 g/100 g (wet basis) moisture content. Sun and Muthukumarappan (2002) found similar results in soy-based extrudates.

The regression equation for the relationship between L value (Y_L) and independent variables in terms of coded variables is presented in Table 3, 2FI model was selected according to Design Expert 6.0, the “ a ” and “ b ” value did not yield any significant model, so only L was selected as the color variable. ANOVA analysis showed that linear term of protein content, and interaction between protein content and moisture content significantly affected the L value. The response surface plot of L value of extrudates are presented in Fig. 4. They showed that the protein content had a negative impact on L value. But L value increased with decreasing feed moisture when the protein content was at low, and L value also increased with increasing processing temperature. This may happened due to Maillard reaction between amino groups and carbonyl groups, which leads to browning in the extrudates. Low feed moisture and high processing temperature are good candidates for the Maillard reaction (Singh et al., 2007).

Table 4
Results of optimization by desirability function based on products served in a liquid.

Run	Constraints	Protein content, P (g/100 g, db)	Moisture content, M (g/100 g)	Temperature, T (°C)	BD (g/ml)	WSI (g/100 g)	RR (g/100 g)	Desirability
1	Min BD	0.61 ((56.1)	−0.16 (39.2)	0.74 (175)	0.42	3.51	135.84	1
		0.49 (54.9)	0.10 (40.1)	0.91 (178)	0.42	3.38	137.55	1
		0.30 (53.0)	−0.16 (39.8)	0.82 (176)	0.42	3.38	144.09	1
2	Max WSI	1.00 (60.0)	1.00 (45.0)	−1.00 (140)	0.72	6.77	86.62	0.99
		1.00 (60.0)	0.97 (45.0)	−1.00 (140)	0.72	6.71	88.23	0.98
		1.00 (60.0)	0.76 (43.8)	−1.00 (140)	0.72	6.32	98.18	0.88
3	Max RR	−0.97 (40.3)	0.83 (44.2)	0.99 (180)	0.62	2.95	210.80	1
		−0.98 (40.2)	0.71 (43.5)	1.00 (180)	0.61	2.97	208.91	1
		−0.92 (40.8)	0.90 (44.5)	1.00 (180)	0.62	2.94	210.51	1
4	Min BD + Max WSI	1.00 (60.0)	−1.00 (35.0)	1.00 (180)	0.41	4.96	142.51	0.74
		1.00 (60.0)	−0.98 (35.0)	1.00 (180)	0.41	4.92	142.04	0.74
		−0.21 (47.9)	1.00 (45.0)	−1.00 (140)	0.55	5.60	64.21	0.71
5	Min BD + Max RR	−0.31 (46.9)	0.06 (40.0)	1.00 (180)	0.47	3.20	168.56	0.83
		−0.31 (46.9)	0.06 (40.0)	1.00 (180)	0.47	3.20	168.80	0.83
		−0.31 (46.9)	0.03 (40.0)	1.00 (180)	0.47	3.22	168.41	0.83
6	Max WSI + Max RR	1.00 (60.0)	−0.91 (35.5)	−1.00 (140)	0.86	4.54	180.37	0.62
		1.00 (40.0)	−0.95 (35.7)	−0.98 (140)	0.86	4.51	181.88	0.61
		1.00 (40.0)	−1.00 (35.0)	−0.95 (141)	0.86	4.47	183.89	0.61
7	Max WSI + Max RR + Min BD	1.00 (60.0)	−1.00 (35.0)	1.00 (180)	0.41	4.96	142.51	0.69
		0.23 (52.3)	−1.00 (35.0)	1.00 (180)	0.43	4.53	152.17	0.66
		1.00 (60.0)	0.29 (41.5)	−0.54 (149)	0.63	4.67	120.17	0.49

Table 5
Results of optimization by desirability function based on extruded snack food products.

Run	Constraints	Protein content, P (g/100 g, db)	Moisture content, M (g/100 g)	Temperature, T	BS (N/mm ²)	ER	L	Desirability
1	Min BS	0.97 (59.7)	1.00 (45.0)	−0.98 (140)	0.12	1.44	79.70	1
		0.87 (58.7)	1.00 (45.0)	−1.00 (140)	0.14	1.45	79.88	1
		−0.97 (40.3)	1.00 (45.0)	−1.00 (140)	0.14	1.39	83.78	1
2	Max ER	0.52 (55.2)	−0.18 (39.1)	0.28 (166)	0.74	1.77	79.14	1
		0.56 (55.6)	−0.10 (39.5)	0.29 (166)	0.74	1.77	79.16	1
		0.58 (55.8)	−0.17 (39.2)	0.33 (167)	0.74	1.77	78.92	1
3	Max L	−0.95 (40.5)	−0.94 (35.3)	−0.95 (141)	0.68	1.33	87.78	1
		−0.98 (40.2)	−0.67 (36.6)	−0.98 (140)	0.66	1.37	87.44	1
		−0.86 (41.4)	−0.97 (35.1)	−0.98 (140)	0.68	1.33	87.39	1
4	Min BS + Max ER	1.00 (60.0)	0.60 (43.0)	−0.72 (146)	0.32	1.57	79.01	0.67
		1.00 (60.0)	0.59 (42.9)	−0.74 (145)	0.32	1.56	78.98	0.67
		1.00 (60.0)	−1.00 (35.0)	−0.06 (159)	0.44	1.65	74.98	0.66
5	Min BS + Max L	−1.00 (40.0)	0.98 (44.6)	−1.00 (140)	0.14	1.38	83.89	0.86
		−1.00 (40.0)	0.99 (44.9)	−0.98 (140)	0.14	1.39	83.86	0.86
		−1.00 (40.0)	0.96 (44.6)	−1.00 (140)	0.15	1.39	83.94	0.85
6	Max ER + Max L	−0.54 (44.6)	−0.11 (34.6)	−0.04 (159)	0.74	1.67	83.30	0.75
		−0.71 (42.9)	1.00 (45.0)	−0.57 (149)	0.32	1.51	83.14	0.63
7	Min BS + Max ER + Max L	−0.71 (42.9)	1.00 (45.0)	−0.58 (148)	0.31	1.51	83.14	0.63
		−0.71 (42.9)	1.00 (45.0)	−0.61 (148)	0.31	1.50	83.14	0.63
		−0.71 (42.9)	1.00 (45.0)	−0.61 (148)	0.31	1.50	83.14	0.63

3.7. Optimum extrusion conditions and characterization of response surfaces of physical properties

In this study, the optimization was applied within the experimental range of total protein content, raw material moisture content and barrel temperature for selected dependent variables to be maximized or minimized either independently or in combination. Second-order polynomial models obtained in this study were utilized for each response in order to determine the specified optimum drying condition. Different thematic scenarios based on economical and industrial constraints were considered. After finding the best solution, a graphical method was applied for mapping the optimum conditions range.

Two types of extruded products were considered in terms of recognizing the importance of different physical properties. The first one was extruded cereal flakes or chunk type of product which are normally soaked in milk prior to consumption. The second type of products were those which can be directly consumed as a snack food. Different physical properties were chosen for the two types to optimize the extrusion condition with Design Expert software.

For the products of first type which may be consumed with milk, WSI, RR, BD were used to optimize the process condition. Table 4 provides typical optimum conditions for this type of products. As can be seen from Table 4, BD was first minimized while other parameters were allowed to be within the experimental range (Run 1). The results show for this condition, 0.42 (g/ml) BD, 3.51 (g/100 g) WSI and 135 (g/100 g) RR obtained under the coded operation condition 0.61 (54 g/100 g real) for protein content, −0.14 (34 g/100 g, wb real) for raw material moisture content and 0.74 (169 °C real) for barrel temperature with the maximum desirability value of 1.0. In Run (2), WSI was maximized while keeping other variables in the range. In this constraint, the desirability was 0.99, and in the run (3), RR was maximized and a desirability of 1 was obtained. Run (4) is a combination of maximizing WSI and minimizing BD, with a compromised desirability of 0.74. Run (5) is a combination of maximizing RR and minimizing BD, with a desirability of 0.83. Run (6), and (7) are all mixed results and give lower desirability (0.62–0.69). One can hypothesize the reason for each of the

constraint depending on the situation. BD minimized to avoid very porous product which will soak quickly and become soggy. Likewise RR was maximized to increase the milk uptake. These two obviously contradict each other and hence combination will provide a lower desirability index.

For the products which may be served directly as a snack food, BS, BD and ER were used to optimize the process condition, and optimum conditions are shown in Table 5. As can be seen in the Table 5, BS was minimized while other parameters were allowed to be in the experimental range (Run 1). The results show for this condition, 0.12 (N/mm²) BS, 1.44 ER and 79.7 (L value) were obtained under the coded operation condition 0.97 (56 g/100 g real) for protein content, 1.0 (41 g/100 g, wb real) for raw material moisture content and −0.98 (148 °C real) for barrel temperature with a high desirability of 1. In Run (2), ER was maximized while keeping other variables in the range. In this constraint, again the desirability was 1, and in the Run (3), L was maximized and again a desirability of 1 could be obtained. Run (4) is a combination of maximizing ER and minimizing BS, a compromising situation with a low desirability of 0.67. In Run (5) L was maximized and BS was minimized, with a slightly higher desirability index of 0.86. Run (6) maximized ER and L with an achieved desirability of 0.75 and Run (7) gave an even lower desirability of 0.63.

The above two are couple of hypothetical scenarios. Other possibilities exist like incorporation in to fruit mixes, in cooking preparations, soups, ice creams, etc. The properties that are important in the product must first be considered prior to evaluating the process for optimization.

4. Conclusions

Extrusion processing variables of soy protein isolate content, feed moisture content and processing temperature significantly influenced the physical properties (BS, BD, ER, WSI, RR and color) of the extrudates. BS and BD increased with increasing soy protein isolate content, but at higher protein content in blend resulted in decreasing ER, WSI, RR and L value. Higher feed moisture played a very important role to increase BS, BD and L value and decrease

WSI and RR. ER showed a maximum cap with when considered with the various process variables. Higher extrusion processing temperature showed a dominant effect to increase BS, ER, RR and L value and decrease BD and WSI. The optimum extrusion processing temperature for BS, BD, ER, WSI, RR and color need to be looked at with respect to the intended type of product and physical property desired. This type of study will be useful in identifying desirable operating conditions for targeted extruded products.

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