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Application of Response Surface Methodology (RSM) for the Production and Optimization of Extruded Instant Porridge from Broken Rice Fractions Blended with Cowpea

Danbaba Nahemiah[1, 3, *], Iro Nkama[2, 3], Mamudu Halidu Badau[3]

[1]Food Technology and Value Addition Research Program, National Cereals Research Institute (NCRI), Badeggi, Bida, Niger State, Nigeria
[2]Department of Food Science and Technology, University of Nigeria, Nsukka, Nigeria
[3]Department of Food Science and Technology, University of Maiduguri, Maiduguri, Nigeria

Email address:
dnahemiah@gmail.com (D. Nahemiah)

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Abstract: Instant porridge from low grade broken rice fractions blended with cowpea to enhance protein quantity and quality was developed in this study through extrusion cooking technology. Response Surface Methodology (RSM) and Central Composite Rotatable Design (CCRD) were adopted for the formulation and optimization of the process variables. The main objective was to obtain the optimum level of extruder barrel temperature (X_1), feed moisture level (X_2) and feed cowpea level (X_3) that will produce optimum porridge having appreciably high protein, water dispersibility, lysine content and calorie value. ANOVA indicated significance ($p<0.05$) of the models fitted in describing the relationship between the input and output variables in its natural state. The coefficient of the determination was also greater than 80% and non-significant lack-of-fit test. Numerical optimization results indicated that the optimum input variables were 120°C barrel temperature, 24g/100g cowpea formulation and 20g/100g feed moisture composition which produce 99.02% dispersibility, 24.13g/100g protein, 73.44% carbohydrate and 388.82kcal/100g calorie, and lysine of 5.02mg/g protein. These data will sharpen the weaning food industry by providing wide opportunity for new food production using low grade rice and common legumes, thereby increasing the economic values of both locally grown rice and cowpea.

Keywords: Rice, Cowpea, Response Surface Methodology, Optimization, Instant Porridge

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1. Introduction

In many sub-Saharan African countries including Nigeria, the level of broken grains obtained after rice milling using the village mills (Engelberg) can be as much as 40% or more (Manful et al., 2004). Such poor-quality rice is often traded at a discount price, reducing the potential margins that could be obtained by rice value-chain actors (Gannon, 1997). The relatively poor quality of locally milled rice in Nigeria and indeed in most of the African countries is one of the primary constraints to further development of the sector (WARDA, 2003). Experts believe that revitalizing the rice sector in terms of both increasing local capacity to compete with imported rice and enhancing value of low quality broken rice through the production of rice-based value added products will improve the market share of locally produced rice greatly (Erenstein and Lancon, 2003).

But, nutritionally, rice and rice-based products are deficient in lysine, an essential amino acid, which can be improved by blending rice with food materials rich in lysine. Food legumes have been proven to be comparatively rich in lysine and therefore combination of rice protein and legume protein provide an ideal source of dietary protein for humans. The utilization of locally grown crops for the production of high protein, shelf stable and affordable recipes in less developed countries has been stressed by international agencies as one of the most suitable channel for addressing the deepening global nutrition challenges (Iwe et al., 2001). Nutritious foods to meet these requirements can be best made from mixture of locally grown cereals and legumes using processing technology that result in shelf stable, convenient and consumer acceptable products using environmentally sustainable technology (Iwe, et al. 2001, Nkama and Filli, 2006; Filli and Nkama, 2007) such as extrusion cooking.

Extrusion cooking technology is a continues high temperature short time (HTST) food processing techniques, in which mechanical energy is combined with heat energy to gelatinize starch and denature proteins, plasticizing and reorganizing food materials to create new shaped and textured products, and also has the ability to inactivate enzymes, destroy some toxic substances and reduce microbial activity (Ryu *et al.*, 1993; Abd El-Hady *et al.*, 1998; Ding *et al.*, 2005). It has been used in the cereal industry for several years to produce many foods and food ingredients such as breakfast cereals, snack foods, baby foods, pasta products, extruded bread, modified starches, beverages, powders, meat and cheese analogues, textured vegetable protein, and blended foods such as corn starch and grounded meats (Anderson *et al.*, 1969; Moore *et al.*, 1990; Abd El-Hady *et al.*, 1998; Zang and Hoseney, 1998; Rhee *et al.*, 1999). It is a technology with high versatility and efficiency, low cost, high output per unit time and short reaction time, with relatively no waste generated (Nabeshima and Grossmann, 2001). During extrusion process, chemical modifications and structural changes occurs in the raw materials, such as starch gelatinization (Akdogan, 1999, Van den Einde *et al.*, 2004), protein denaturation (Iwe *et al.*, 2004), pigment and vitamin degradation (Ilo and Berghofer, 1999), and loss of volatile compounds (Bhandani *et al.*, 2001). These changes resulted in new food product with new functional, nutritional and sensory qualities (Bryant, *et al.*, 2001). The knowledge of changes in extruder operating variables therefore provide necessary information for the prediction of what fraction of food materials will undergo specific reaction during extrusion process and its possible effects on the quality of finished product. Ding *et al.*, (2005) reported that little change in extrusion variables such as feed compositions, feed moisture content, screw speed, screw geometry, die configuration; feed rate, processing temperature could greatly affect finished product quality. This therefore has placed a critical need on food scientists to properly and effectively optimize production variables if extrusion technology is to be adopted.

Response surface methodology (RSM) and central composite design (CCD) provides an ideal tool for investigating and optimizing process and product parameters in food processing. RSM is a collection of mathematical and statistical techniques that are useful for modeling and analyzing of problems in which a response of interest is influenced by several variables and the objective is to optimize this response (Montgomery, 1997; Noordin *et al.*, 2004). Several workers (King and Zall, 1992; Filli *et al.*, 2011 and 2012) have used RSM to predict optimum food processing conditions. Optimal process and product design will further improve efficient utilization of broken rice in the production of high quality nutritious products, reduce production cost, and hasten the upscale of these products from pilot status to industrial scale and also facilitate easy troubleshooting and quality control at large scale. Optimization of extrusion cooking therefore, may involves critical consideration of process parameter, system parameter and product quality.

The current study focuses on the optimization of extrusion parameters in the use of broken rice grain and cowpea for the production of value-added instant porridge using response surface methodology and central composite design.

2. Materials and Methods

2.1. Materials

Broken rice fractions from the milling of FARO 52 obtained from 2013 rain-fed production were obtained from the National Cereals Research Institute (NCRI) Badeggi, Nigeria, and cowpea (local variety) purchased from the modern market, Bida, Niger State, Nigeria. All samples were manually cleaned and packaged in a sealed polyethylene bags before storage in a cardboard at ambient temperature (32±2°C)

2.2. Methods

Broken rice fractions were blended at different proportion with cowpea flour according to response surface methodology experimental design and extruded at different combinations of extruder barrel temperature and blend moisture composition. The extrudates were pulverized and reconstituted with water. Optimum nutritional composition was determined using standard procedure.

2.3. Cowpea Flour Preparation

Cowpea flour was prepared by first steeping 30kg in clean tap water for 30min at room temperature (32±2°C) and dehulled by gently pounding in wooden mortar and pestle followed by several washing and final drying to about 10-12% moisture levels. Dried cotyledons were then milled in an attrition mill (Locally fabricated) and sieved with fine (150µm) laboratory sieve (Brabender OHG Duisburg). Samples were then packaged in a sealed polyethylene bags at room temperature until required.

2.4. Rice Flour Preparation

Broken rice were manually sorted to remove impurities and grounded in an attrition mill (Locally fabricated) and sieved with fine (150µm) laboratory sieve (Brabender OHG Duisburg). Samples were then packaged separately in a sealed polyethylene bags at room temperature until required.

2.5. Blend Formulation and Moisture Adjustment

Fifteen (15) formulations were prepared to contain cowpea flour (wet basis) ranging between 8 - 24% based on the experimental layout in Table 2. The blended samples were conditioned to appropriate moisture content by spraying with a calculated amount of water and mixing continuously at medium speed in a blender (Laboratory Scale Hobart Mixer, Hobart Corporation, Troy, Ohio, USA). The samples were put in closed plastic buckets and stored overnight (32±2°C). The feed materials were then allowed to stand for 3hrs to equilibrate at room temperature prior to extrusion exercise. The amount of water added was calculated using the equation earlier proposed by Ascheri, 2010.

$$\text{Amount of water to be added (g)} = \frac{(M_f - M_i)x\, S_w}{100 - M_f} \quad (1)$$

Where M_f = Final moisture content, M_i = Initial moisture content and S_w = Sample weight (g)

2.6. Extrusion Cooking Processing

The extrusion processing was performed using a co-rotating twin-screw (SLG 65 Twin-Screw Extruder, Jinan Saibainuo Technology Development Company Ltd, Peoples Republic of China) extruder with an operating screw speed range of 0 to 300rpm, length to diameter ratio of barrel was 20:1, while the diameter of the screw was 30mm. The formulations were introduced manually into the feeding zone through a conical fed hopper at the rate of 30kg/h while avoiding accumulation of fed material, where it first got gelatinized and plasticized under thermal and mechanical energy generated by the double screws; the paste then was cooked and extruded through the die. At a steady state, samples were collected and processed for analysis. The barrel temperature ranges were between 84 – 140°C, and feed moisture content was set at 15-24%.

2.7. Experiment Design and Statistical Analysis

Table 1. *Independent variables and natural levels used for Central Composite Rotatable Design.*

Independent variables	**Levels of coded variables**				
	-α	**Low**	**Medium**	**High**	**+α**
	-1.68	**-1**	**0**	**1**	**+1.68**
Barrel Temperature (X_1)	86.36	100	120	140	153.64
Feed Moisture content (X_2)	11.59	15	20	25	28.41
Feed Composition (X_3)	2.55	8	16	24	29.45

Level of each variable was established based on a preliminary extrusion. The distance of the axial points from the centre point was ± 1.68, and calculated from Equation $\alpha = (2^n)^{1/4}$ where n is the number of variables.

The extrusion conditions were optimized with a three-factor five-level central composite rotatable design (CCRD) (Box and Hunter, 1957). Response Surface Methodology (RSM) was used to investigate the effect of the independent variables on the responses. Extruder barrel temperature (X_1), feed moisture content (X_2) and formulation cowpea composition (X_3) were the independent variables considered and the qualities of the finished products were the response variables measured. In order to objectively define the experimental ranges, preliminary experiments were conducted to establish the narrower, more effective ranges of the independent variables (X_1, X_2, and X_3) prior to the experimental runs. As the design value ranges were established, they were coded to lie at ±1α for the factorial points, 0 for the center points and ±1α for axial points. The codes were calculated as a function of the range of interest of each factor as presented in Table 1. The experiments were randomized to maximize the effects of unexplained variability in the observed responses due to extraneous factors, while five replicates at the center of the design were used to allow for estimation of pure error sum of square and lack-of-fit. Analysis of variance (ANOVA) was conducted to determine significant differences among the mean treatment combinations.

Table 2. *Outline of experimental design with variables in their coded and un-coded forms.*

Experimental Runs	**Coded and un-coded Independent variables**		
	X_1 (°C)	**X_2 (g H_2O/g Sample)**	**X_3 (g/100g Sample)**
1	-1(100)	-1(15)	-1(8)
2	1(140)	-1(15)	-1(8)
3	-1(100)	1(25)	-1(8)
4	1(140)	1(25)	-1(8)
5	-1(100)	-1(15)	1(24)
6	1(140)	-1(15)	1(24)
7	-1(100)	1(25)	1(24)
8	1(140)	1(25)	1(24)
9	-1.68 (86..4)	0(20)	0(16)
10	1.68 (153.6)	0(20)	0(16)
11	0(120)	-1.68 (11.6)	0(16)
12	0(120)	1.68 (28.4)	0(16)
13	0(120)	0 (20)	-1.68(2.6)
14	0(120)	0(20)	1.68(29.5)
15	0(120)	0(20)	0(16)

X_1 = Barrel temperature, X_2 = Feed moisture content, X_3 = Feed cowpea composition. Duplicate runs were carried out at all design point except at the center point where five measurements were carried out and average recorded. The experimental runs were randomized.

2.8. Process Optimization

A second order polynomial regression equation was modeled on the basis of the experimental data and optimum parameters defined using Matrix Laboratory (MATLAB 14.13) Software. From the resulting values, for each of the response variable, the coefficients of the polynomial equation (β_o, β_i and β_{ij}) are determined and the equation simplified based on the influence of the factors on the final response. The responses were then expressed as second-order polynomial equation according to Eq. 2.

$$\mathrm{Y} = f(y) = \beta_0 + \sum_{i=1}^{k} \beta_i X_i + \sum_{i=1}^{k} \beta_{ii} X_i^2 + \sum_{i=1}^{k} \sum_{i=1}^{k} \beta_{ij} X_i X_j + \varepsilon \quad (2)$$

Where Y is the predicted response used as a dependent variable, *k* is the number of independent variables considered in the experiment; β_o constant coefficient and β_i, β_{ij} and β_{ii} are the coefficient of linear, interaction and square terms respectively, while ε is the random error term. Multivariate regression analysis with model equation (2) was carried out on the data using MINITAB 14.13 statistical software (Manitab Inc. USA) to yield equation 3 which was used to optimize the product responses (Filli et al, 2011).

$$Y = \beta_o + \beta_1X_1 + \beta_2X_2 + \beta_3X_3 + \beta_{11}X^2{}_1 + \beta_{22}X^2{}_2 + \beta_{33}X^2{}_3 + \beta_{12}\,X_1X_2 + \beta_{13}\,X_1X_3 + \beta_{23}\,X_2X_3 + \varepsilon \qquad (3)$$

The outline of the experiments with the coded and natural values are presented in Table 2 and to provide homogeneous variance as required for regression models, the data were first transformed to standard scores ($z = \frac{x-X}{s}$) where x is the dependent variable of interest, X mean of the dependent variables and s standard deviation.

2.9. Validation of Fitted Models

To check if the fitted models provide an adequate approximation to the real system, it is often important to check model adequacy. Unless a model shows adequate fit, proceeding with optimization may lead to misleading results. In this study, numerical validations were conducted on the fitted models. Numerical methods involve the analysis of the coefficient of determination (R^2) and adjusted coefficient of determination (R^2_{adj}) calculated as:

$$R^2 = 1 - \frac{\text{SS residual}}{\text{SS model+SS residual}} \qquad (4)$$

$$Adjusted\ R^2 = 1 - \frac{\text{n}-1}{\text{n}-\text{p}}\left(1 - R^2\right) \qquad (5)$$

Where SS is the sum of squares, n is the number of experiments and p is the number of predictors in the model not counting the constant term. R^2 value close to unity and R^2_{adj} close to R^2 ensure satisfactory fitting of the model to the real system. Probability value (p-value) was also used to check for the significance of each factor and interaction between the factors. The smaller the p-value, the more significant is the corresponding coefficients (Mason et al. 2003). Since triplicate measurements were recorded during analysis of the response variables, a lack-of-fit test was conducted to examine the significance of replicate error in comparison to the model dependent error. This test split the residual or error sum of squares into two parts, one due to pure error as a result of duplicate measurement and the second due to lack-of-fit which is the ratio between the lack-of-fit mean square and pure error mean square. F-test can then me used to measure wither the lack-of-fit is statistically significant or not at the described level of probability. Non-significant lack-of-fit was considered desirable.

2.10. Dispersibility Measurement

Dispersibility was determined by placing 15g of sample in a 100ml measuring cylinder and adding water to make up to volume, then stir for 1min 30sec and allowing it to settle for 15min. The volume of the settled particle was subtracted from 100 and the difference reported as dispersibility in percentage.

2.11. Proximate Composition

Protein, fat, ash, dietary fiber, mineral (magnesium, manganese, calcium, zinc, copper and iron) and were determined according to AOAC method (1984). The percentage carbohydrate was calculated by difference. Gross energy value (Kcal/100g) was calculated (FAO, 2002).

2.12. Amino Acids Profile Analysis

Amino acids were determined according to methods described by Onyeike et al (2005) as modified by Anyalogbu et al (2015). Five (5g) of sample was weighed into extraction thimble of Soxhlet extraction apparatus and 60ml of chloroform/methanol (2:1 v/v) for extraction. This was followed by hydrolysis by weighing the defatted sample into a glass ampoule and 7ml of 6MHCl added. The glass ampoule was then sealed with flame from Bunsen burner before incubation at 105±2°C for 22hr to effect hydrolysis (Nwonsu, et al., 2008). After incubation, the ampoule was allowed to cool to ambient temperature (32±2°C) before opening at the tip and the content filtered through filter paper to remove humis. The protein hydrolysate was evaporated to dryness at 40°C under vacuum in a rotary evaporator (Buchi Rotavopour Switzerland) and the residual dissolved with 5ml acetate buffer (pH 2.0), in a specimen bottle and stored in a freezer for analysis. 10ml of the dissolved residual was collected with a micro-syringe and dispensed into the cartridge of the TSM amino acid analyser. The amino acids were separated on the ion-exchange column through a combination of change in pH and cation strength. The post column reaction between ninhydrin and amino acids eluted from the column formed Riemann's purple, a diketohydrinhylidene-diketohydrindamine (Friedman, 2004). The reaction was therefore monitored at 440nm and 570nm wavelengths. It took about 1hr, 15 minutes to complete the reaction. Net height of each peak produced by the chart recorder of TSM (each representing an amino) was measured. The half-height of the peak on the chart was found and width of the peak on the half height was accurately measured and recorded. Approximately area of each peak was then obtained by multiplying the height with the width at half-height. Norleucine was added as internal standard and a standard amino acid mixture (Beckman No. 338088, Beckman Coulter, CA) was measured under the same condition as sample. Finally, the amount of each amino acid present in the sample was calculated as g/100g protein. The amino acids, leucine, isoleucine, lysine, methionine, phenylalanine, threonine, tryptophan, valine, tyrosine and tryptophan were determined.

3. Results and Discussion

3.1. Model Fitting and Validation

This study was carried out to produce nutritious extruded instant rice-cowpea porridge and to optimize the production process variables and product quality using response surface methodology and central composite design. Process variables considered were barrel temperature (X_1), feed moisture content (X_2) and feed cowpea composition (X_3), while the response variables were dispersibility index, proximate composition and amino acids composition. The mean response values of proximate composition and amino acids profile are presented in Tables 3 and 4 respectively. The independent and response variables were fitted to the second-order model equation (Eq. 2) and its goodness of fit examined using analysis of variance (ANOVA), coefficient of determination (R^2 and R^2_{adj}), lack-of-fit test and analysis of residual. The predictive regression models developed for the relationship between the dependent (y) and independent (X) in terms of proximate composition of extruded instant rice-cowpea porridge is presented in Eq. 6 to 12 for moisture, lipid, protein, ash, carbohydrate and caloric value respectively . The coefficients with single factor (X_1, X_2, and X_3) represent the independent effect of a particular variable, while coefficients with two of the factors (X_1X_2, X_1X_3, and X_2X_3) and the ones with second-order terms (X_1^2, X_2^2, and X_3^2) represent interaction between the three factors and quadratic effects respectively. A positive sign in front of the regression term is an indication of synergetic relationship, while negative sign indicates an antagonistic relationship.

Moisture

$$5.49148 – 0.07059X_1 – 0.05258X_2 - 0.01937X_3 + 0.00033X^2{}_1 + 0.0027X^2{}_2 + 0.00093X^2{}_3 – 0.00044\,X_1X_2 – 0.00005\,X_1X_3 – 0.00072\,X_2X_3 \tag{6}$$

Lipid

$$6.85183 – 0.06165X_1 – 0.17541X_2 – 0.14678X_3 + 0.0002X^2{}_1 + 0.00272X^2{}_2 + 0.00138X^2{}_3 + 0.00032\,X_1X_2 + 0.00043\,X_1X_3 + 0.0017\,X_2X_3 \tag{7}$$

Protein

$$-98.7103 + 1.5581X_1 + 3.7519X_2 + 0.1058X_3 – 0.0073X^2{}_1 – 0.0736X^2{}_2 – 0.0195X^2{}_3 – 0.0018\,X_1X_2 + 0.0116\,X_1X_3 – 0.0436\,X_2X_3 \tag{8}$$

Fibre

$$8.90382 – 0.04286X_1 – 0.47754X_2 + 0.04467X_3 + 0.00026X^2{}_1 + 0.00999X^2{}_2 + 0.00191X^2{}_3 + 0.00029\,X_1X_2 – 0.00143\,X_1X_3 + 0.00203X_2X_3 \tag{9}$$

Ash

$$0.262198 + 0.00427X_1 – 0.02886X_2 + 0.080508X_3 – 0.000043X^2{}_1 – 0.000898X^2{}_2 – 0.000572X^2{}_3 + 0.000656\,X_1X_2 – 0.00034X_1X_3 – 0.000891\,X_2X_3 \tag{10}$$

Carbohydrate

$$182.284 – 1.459X_1 – 3.051X_2 – 0.065X_3 + 0.007X^2{}_1 + 0.062X^2{}_2 + 0.017X^2{}_3 – 0.010\,\beta_{13}\,X_1X_3 + 0.040\,X_2X_3 \tag{11}$$

Calorie value

$$397.783 – 0.160X_1 + 1.143X_2 – 1.207X_{3}\text{-}\ 0.022\beta_{22}X^2{}_2 + 0.001X^2{}_3 – 0.002\,X_1X_2 + 0.009X_1X_3 + 0.003\,X_2X_3 \tag{12}$$

While the predictive models representing the relationship between the extruder (process) variables and the response variables in terms of amino acids profile of the instant porridge are outlined in the regression models equation 13 to 20. The results of ANOVA performed on the models to evaluate the significance of the linear, quadratic and interactive effects of the independent variables on the dependent variables are presented in Tables 5 and 6 respectively for proximate and amino acid profiles respectively. This analysis was done using the Fisher's F-test. In this study, the ANOVA indicated significant ($p<0.05$) models since probability value is less than 0.05. The coefficients of determination (R^2 and R^2_{adj}) results also shows that the proximate composition predictive models R^2 values ranged between 59.00 and 99.5% (Table 5) and R^2_{adj} varying between 40.6 and 99.5% (Table 6). R^2 is the ratio of the explained variation to the total variation and measures the degree of fitness of a regression model (Singh et al. 2007; Filli et al. 2011); it therefore defines the proportion of the variability in the observed response variables which is accounted for by regression analysis (McLaren et al., 1977; Filli et al., 2011). The closer the R^2 value is to unity, the better the empirical model fits the actual data (Lee and Wang 1997; Zaibunnisa et al*.,* 2009), but the less the value of R^2, the less relevant the determinant variables in the model have in explaining variability observed in the response variables. Zaibunnisa et al. (2009) suggested that R^2 value should be at least 80% to have good fit of a regression model. The results of this study therefore showed that the model for all the response variables were highly adequate to explain the variability in response because they have satisfactory level of R^2 which is higher than 80% and R^2_{adj} that is close to R^2. But that of moisture content was less than 80% and is likely be as non-significance of some terms added in the model.

As repeated measurements were carried out during the data generation, lack-of-fit test which indicate the significance of the replicate error in comparison with the model dependent error was carried out. This test split the error sum of squares into two portions, one which is due to pure error and other due to lack-of-fit. F-test was then used to determine wither the lack-of-fit test was significant or not. The results indicated non-significant ($p<0.05$) lack-of-fit in both proximate and amino acid profiles (Tables 5 and 6). Non-significant lack-of-fit therefore is desired as a significant test indicates that there may be contributions in the regression response relationship that are accounted for by the fitted models. It is appropriate therefore to conclude that the fitted models adequately approximates the responses and can be used satisfactorily for the prediction of any value of the responses within the defined experimental range.

Lysine

$$-2.6686 + 0.0633X_1 + 0.3289X_2 + 0.0329X_3 – 0.00005X^2{}_1 – 0.0033X^2{}_2 + 0.00014X^2{}_3 – 0.0015\,X_1X_2 - 0.0008\,X_1X_3 + 0.00103\,X_2X_3 \tag{13}$$

Isoleucine

$$-4.5332 + 0.0729X_1 + 0.1826X_2 + 0.1509X_3 - 0.0002X^2{}_1 - 0.0021X^2{}_2 - 0.00104X^2{}_3 - 0.0007\,X_1X_2\ 0.00093\,X_1X_3 - 0.00066\,X_2X_3 \quad (14)$$

Lucien

$$10.5104 - 0.1743X_1 + 0.2673_2X_2 + 0.2458X_3 + 0.0015X^2{}_1 + 0.0103X^2{}_2 + 0.0029X^2{}_3 - 0.0052\,X_1X_2 - 0.0035\,X_1X_3 + 0.0008\,X_2X_3 \quad (15)$$

Valine

$$11.9071 - 0.1714X_1 + 0.0460X_2 + 0.0643X_3 + 0.0008X^2{}_1 + 0.0019X^2{}_2 - 0.0012X^2{}_3 - 0.001\,X_1X_2 - 0.0001\,X_1X_3 - 0.0008\,X_2X_3 \quad (16)$$

Methionine + Cysteine

$$2.3624 - 0.0361X_1 + 0.0357X_2 + 0.0315X_3 + 0.0003X^2{}_1 + 0.0022X^2{}_2 + 0.0009X^2{}_3 - 0.0010\,X_1X_2 - 0.0004\,X_1X_3 + 0.0006\,X_2X_3 \quad (17)$$

Threonine

$$-4.4485 + 0.0758X_1 + 0.1337X_2 + 0.1371X_3 - 0.00015X^2{}_1 - 0.00153X^2{}_2 - 0.00038X^2{}_3 - 0.00079\,X_1X_2 - 0.00137\,X_1X_3 + 0.00153\,X_2X_3 \quad (18)$$

Tyrosine +Phenylalanine

$$7.0172 - 0.0676X_1 + 0.0539X_2 - 0.0875X_3 + 0.0004X^2{}_1 - 0.0008X^2{}_2 + 0.0025X^2{}_3 - 0.0004\,X_1X_2 - 0.0007\,X_1X_3 + 0.0040\,X_2X_3 \quad (19)$$

Tryptophan

$$12.4167 - 0.1881X_1 - 0.0162X_2 + 0.0003X_3 + 0.0009X^2{}_1 + 0.0036X^2{}_2 + 0.0017X^2{}_3 - 0.0012\,X_1X_2 - 0.0008\,X_1X_3 + 0.0018\,X_2X_3 \quad (20)$$

3.2. Instantization of Rice-Cowpea Porridge

Instant porridges are precooked flakes which are readily reconstitutable upon the addition of water or milk to yield product having cooked texture. This is one of the functional qualities of food products and has significant impact on consumer acceptability of a product because of its convenience in preparation. In this study, the dispersibility of the porridge in water was evaluated to establish it instant reconstitution upon the addition of water. The ability of food particle to be wet without the formation of lumps, with simultaneous disintegration of agglomerates is an indication of its reconstitution in water to give a fine and consistent paste (Kulkarni et al. 1991; Eke-Ejiofor and Owuno, 2012; Otegbayo et al. 2013). Therefore, high dispersibility is required for a food to be considered instant. In this study, the dispersibility of the product (not presented in the Tables) ranged between 98.2 to 99.2% indicating high reconstitution. These values are higher than 63.0 to 87.0% earlier reported by Otegbayo et al. (2013). Pulverized food materials are 'instantized' when the surface of each particle is easily wetted and the material rehydrated and particles sink below the surface of the solvent to disperse rapidly through the solvent. This attributes are termed wettability, sinkability, dispersibility and solubility (Filli et al. 2011). For a product to be considered instant therefore, it most completes these stages within a few second. The porridge in this study may therefore be considered an instant porridge.

3.3. Effects of Processing Variables on Proximate Composition

The results of mean observed values for proximate composition (moisture, protein, lipid, fibre, ash, carbohydrate and caloric value) for rice-cowpea porridge is presented in Table 3. The moisture content ranged between 0.08 and 0.92% with an average of 0.51%. The highest moisture content was recorded in sample corresponding to barrel temperature 100°C, moisture 20% and feed composition 16% and the least value in sample extruded at 120°C barrel temperature, 20% moisture and 16% cowpea content, the moisture content increase with decreasing moisture content. It is also clear from these results that moisture content of extruded sample depend mainly on the extrusion temperature and not on the amount of moisture of raw material or composition of cowpea. These results suggested that the product had low moisture enough to have an extended shelf life. It has also been observed by several authors that in a dry food systems with moisture content between 6% and 10%, there is a prolong shelf stability, and above this range, the stability of the system could be impeded by both chemical and microbiological agents (Harper and Jansen, 1985; Afoakwa et al. 2006; Asare et al. 2012). The protein content increased from 19.00 to 29.06% representing about 65.38% increase when temperature drops from 140°C to 120°C and when feed moisture and cowpea content of feed increased from 15 to 20% and 8 to 16% suggesting proportional increase in protein when rice is fortified with cowpea. Filli et al (2011) reported an increase from 11.23 to 16.23 protein content when millet was fortified with cowpea through extrusion cooking. Protein content of foods are determined as nitrogen multiply by a factor (Nx6.25), and apparent protein content is not affected by extrusion temperature as nitrogen is not affected by heat (Pelembe et al., 2002; Filli et al., 2011). The fat content ranged between 0.04 and 0.61%, with the highest value observed in sample extruded at 100°C barrel temperature, 15% moisture and 8% cowpea. The increase in fat content may be attributed to the possible raising concentration of some non-fat compounds formed by Millard reaction and/or caramelization which are insoluble in organic solvent (El-Samahy et al., 2007). The significantly low fat content implies that this quality parameter needs to be added from other sources into the diet of the consumers of this product especially if it is going to be used as weaning food. Mitzner and co-workers (1984) reported that for a food to be used as complementary formulations, the minimum fat content requirement should be 6%. Both rice and cowpea are low in fat content, in addition, the extruder processing condition affect the little fat present in the raw materials through several reactions including complex formation with amylose and protein, oxidation, lipid binding due to interactions with starch and protein's cis and trans isomerization of unsaturated fatty acids and degradation of fat splitting enzymes. All these reactions resulted in fat loss. The fibre contents varied between 1.62 to 2.88%, with the least value recorded in sample extruded at 120°C barrel temperature, 20% moisture and 29.5% cowpea and the highest value observed in sample corresponding to 140°C barrel temperature, 15% moisture and 8% cowpea extrusion conditions. During extrusion cooking process, positive effects of the process parameters on the total and soluble fiber has been observed (Rashid, et al., 2015). Insoluble dietary fiber decreased apparently as the process parameters changes. These changes may probably be due to disruptions of covalent and non-covalent bonds in the carbohydrate and protein moieties leading to smaller and more soluble molecular fragments. The ash content which is the indication of mineral content of the extruded instant porridge varied between 0.80 and 1.10%, with the highest value observed in sample corresponding to 140°C barrel temperature, 25% moisture and 8% cowpea extrusion conditions and the least value seen in sample extruded at 100°C barrel temperature, 25% moisture and 8% cowpea. These results is contrary to an earlier observation by El-Samahy et al., (2007), who observed significant increase in ash content when cactus pear concentration was increased in a rice-cactus pear extruded samples. With respect to the carbohydrate and calorie values of the rice-cowpea extrudates, the highest values were 72.21% and 391.01kcal/100g in samples 10 (153.6°C barrel temperature, 20% moisture and 16% cowpea) and 1 (100°C barrel temperature, 15% moisture and 8% cowpea) respectively, while the least values were observed in samples 15 (120°C barrel temperature, 15% moisture and 24% cowpea) and 5 (100°C barrel temperature, 15% moisture and 24% cowpea) respectively.

Regression coefficients (β - this is the mean change in the response variables for a unit change in the independent variables while holding the other predictors in the model constant) of the effects of the barrel temperature, feed moisture content and feed blend composition on the proximate composition indicated negative linear and interactive effects of these variables on the moisture content (Table 5) and the ANOVA indicates that both linear and quadratic effects were significant ($p<0.05$). This implies that increasing the cowpea content may linearly increase the moisture content of the resulting extruded instant porridge. The protein content coefficient of regression also shows that there were significant ($p<0.05$) linear, quadratic and interactive effects of the process variables on the protein results (Table 5). The negative linear effect suggests that increasing feed cowpea composition, barrel temperature and feed moisture content resulted in decreased protein content in the porridge, while the significance of the linear, quadratic and interactive terms indicated that these changes may not be attributed to a single factor alone. Though, it has been established that increasing protein based component of feed material directly increase product protein content (Phillips et al. 1984; Singh et al. 1991), but because of the effects of high temperature and low moisture content during extrusion, the protein content were reduced due to denaturation. Though the protein content were significantly affected, at a steady state, the protein content produced in this study is above the minimum protein level of 15.7% recommended by FAO/WHO/UNN (1985) for supplementation feeding. The fat content were significantly ($p<0.05$) affected by the linear, quadratic and interactive terms, with the quadratic and linear in an antagonistic manner (Table 5), while the ash content were negatively affected by the process variables. These results is contrary to an earlier findings by Obatolu (2002) and Filli et al., (2011) who observed positive relationship between ash content and feed moisture content. Though the ash content is generally low as observed by this workers in cereal/legume extruded formulations, this may likely be as a result of low ash in dehulled beans and polished rice. The calorie content of the porridge was significantly ($p<0.05$) affected by the process variables and the regression coefficient results indicated synergetic linear effect of feed moisture content and antagonistic effects of feed cowpea composition and barrel temperature.

3.4. Effects of Processing Variables on Amino Acid Profile

The mean amino acid composition as affected by the processing parameters is presented in Table 4. The lysine content of the rice-cowpea based porridge recorded the lowest value of 4.12g/100g protein representing 120°C barrel temperature, 20% feed moisture composition and 2.5% cowpea in the blend, while the highest value of 6.00g/100g protein was observed in sample representing experimental condition of 120°C extrusion temperature, 20% feed moisture composition and 16% feed cowpea composition (Table 4), this value are higher than the 340mg/gN recommended by WHO as recommended dietary allowance (RDA). For the isoleucine content of the extruded rice-cowpea formulations, the highest value of 3.20 was recorded in experimental run 2, representing 140°C, 15% and 8% barrel temperature, feed moisture and blend composition experimental conditions respectively and the lowest result of 2.41g/100g protein was observed in run 5 (100°C barrel temperature, 15% feed moisture and 24% cowpea composition). The leucine content of rice-cowpea based extrudates ranged from 6.45g/100g protein for design point 5 (100°C barrel temperature, 15% moisture content and 24% feed composition) to 10.74g/100g protein in design point 4 processed at 140°C barrel temperature, 25% feed moisture content and 8% blend composition (Table 4). It is also clear from the results in Table 4 that the valine content varied between 2.15g/100g protein corresponding to design point 14 and extruded at extruder condition of 120°C barrel temperature, 20% feed moisture content and 16% cowpea composition, while the highest value of 3.89g/100g protein was recorded in run 4 which correspond to barrel temperature of 140°C, feed moisture composition of 25% and 8% cowpea composition extrusion conditions.

With regard to methionine + cysteine, the value varied between 1.99 and 2.99g/100g protein, with the lowest value recorded in design 5 and the highest in design 4 which corresponds to extrusion conditions of 100°C barrel temperature, 15% moisture content, 24% blend composition and 140°C barrel temperature, 25% feed moisture content and 8% blend cowpea composition respectively (Table 4). The threonine composition of rice-cowpea based extruded ranged between 2.32 and 2.95g/100g protein as recorded in designs 14 and 4 respectively. The lowest value was obtained when the extrusion conditions were at 120°C barrel temperature setting, 20% feed moisture and 16% feed blend composition, while the highest value was recorded at 140°C extruder barrel temperature, 25% feed moisture and 8% cowpea composition. While the tyrosine + phenylalanine ranged from 5.28g/100g protein in design 11 corresponding to 120°C barrel temperature, 11.6% feed moisture content and 16% feed cowpea composition to 8.08g/100g protein in design point 4 representing 140°C barrel temperature, 25% feed moisture composition and 8% cowpea composition. For the tryptophan content, design point 5 corresponding to 100°C barrel temperature, 15% feed moisture content and 24% cowpea composition recorded the lowest value of 1.02g/100g protein, while design point 4 corresponding to 140°C barrel temperature, 25% feed moisture and 8% cowpea composition recorded the highest value of 1.61g/100g protein (Table 4).

Table 3. *Mean proximate composition of extruded instant rice-cowpea porridge as affected by extrusion process.*

Independent variables[a]			Response variables[b]						
X1	**X2**	**X3**	**Moisture (%)**	**Protein (%)**	**Fat (%)**	**Fiber (%)**	**Ash (%)**	**CHO (%)**	**Calories (Kcal/100g)**
-1(100)	-1(15)	-1(8)	0.68	24.51	0.61	2.11	0.90	71.97	391.01
1(140)	-1(15)	-1(8)	0.70	19.00	0.40	2.88	0.92	76.80	386.80
-1(100)	1(25)	-1(8)	0.76	27.36	0.40	2.01	0.80	69.43	390.76
1(140)	1(25)	-1(8)	0.59	21.20	0.30	2.46	1.10	74.94	387.26
-1(100)	-1(15)	1(24)	0.59	24.10	0.05	2.31	1.11	72.44	386.61
1(140)	-1(15)	1(24)	0.50	26.11	0.11	1.70	0.96	71.12	389.91
-1(100)	1(25)	1(24)	0.41	20.08	0.10	2.11	0.91	76.80	388.42
1(140)	1(25)	1(24)	0.10	21.26	0.30	2.08	0.99	75.37	389.22
-1.68 (86..4)	0(20)	0(16)	0.92	22.58	0.31	2.00	0.89	74.22	389.99
1.68 (153.6)	0(20)	0(16)	0.66	19.52	0.26	2.01	1.00	77.21	389.26
0(120)	-1.68 (11.6)	0(16)	0.57	25.11	0.28	2.56	0.90	71.15	387.56
0(120)	1.68 (28.4)	0(16)	0.30	23.10	0.25	2.31	0.96	73.38	388.17
0(120)	0 (20)	-1.68(2.6)	0.59	25.63	0.51	2.51	0.83	70.52	389.19
0(120)	0(20)	1.68(29.5)	0.19	25.99	0.09	1.62	0.96	71.34	390.13
0(120)	0(20)	0(16)	0.08	29.06	0.04	1.73	1.02	68.15	389.20

[a]X_1 = barrel temperature (°C), X_2 =feed moisture content (%), X_3 = feed blend composition (%), CHO = carbohydrate. [b]Values are mean of triplicate determinations, except at the centre points where five readings were taken and average recorded

Table 4. *Mean amino acid composition of extruded instant rice-cowpea porridge as affected by extrusion process.*

Independent variables[a]			Response variables[b]							
X_1	**X_2**	**X_3**	**Lysine**	**ISOLEU**	**Leusine**	**Valine**	**Meth + Cysteine**	**THRE**	**TYRO+PHEN**	**TRYP**
-1(100)	-1(15)	-1(8)	4.84	2.67	7.10	3.28	2.29	2.44	6.70	1.18
1(140)	-1(15)	-1(8)	5.44	3.20	9.98	3.77	2.74	2.90	7.80	1.50
-1(100)	1(25)	-1(8)	5.11	3.00	8.69	3.53	2.45	2.55	6.85	1.34
1(140)	1(25)	-1(8)	5.61	3.20	10.74	3.89	2.99	2.95	8.06	1.61
-1(100)	-1(15)	1(24)	4.45	2.41	6.45	2.98	1.99	2.33	5.66	1.02
1(140)	-1(15)	1(24)	5.06	2.80	7.80	3.47	2.40	2.44	6.85	1.29
-1(100)	1(25)	1(24)	5.39	3.07	9.27	3.65	2.69	2.78	7.54	1.45
1(140)	1(25)	1(24)	4.89	2.67	7.51	3.41	2.34	2.44	7.03	1.23
-1.68 (86..4)	0(20)	0(16)	4.78	2.61	6.81	3.22	2.18	2.38	6.17	1.07
1.68 (153.6)	0(20)	0(16)	5.22	3.07	9.10	3.59	2.50	2.61	7.45	1.39
0(120)	-1.68 (11.6)	0(16)	4.56	2.78	7.11	3.00	2.33	2.44	5.28	1.12
0(120)	1.68 (28.4)	0(16)	5.13	2.88	6.78	2.17	2.15	2.51	5.78	1.06
0(120)	0 (20)	-1.68(2.6)	4.12	2.79	6.77	2.18	2.15	2.52	6.02	1.05
0(120)	0(20)	1.68(29.5)	6.00	2.77	6.79	2.15	2.14	2.32	6.10	1.06
0(120)	0(20)	0(16)	5.14	2.98	6.98	3.00	2.13	2.41	5.82	1.07

ISOLEU = Isoleucine, Meth = Methionine, THRE = Threonine, TRYO = Tryosine, PHEN = Phenylalanine, TRYP = Tryptophan, X_1 = Barrel temperature, X_2 = Feed moisture content, X_3 = Feed composition. [b]Values are mean of triplicate determinations, except at the centre points where five readings were taken and average recorded. The experimental runs were randomized

Table 5. *Estimated regression equation coefficients for response variables (proximate composition) in rice-cowpea instant porridge.*

Term	Moisture	Protein	Fat	Fiber	Ash	CHO	Calories
Linear							
X1	-0.07059**	-0.06165**	1.5581**	-0.04286	0.0043	-1.459**	-0.160
X2	-0.05258	-0.17541**	3.7519**	-0.47754**	-0.0289	-3.051**	1.143**
X3	-0.01937	-0.14678**	0.1058	0.04467	0.0805**	-0.065	-1.207**
Quadratic							
X12	0.00033**	0.00020**	-0.0073**	0.00026**	-0.00004	0.007**	<0.0001
X22	0.00270	0.00272**	-0.0736**	0.00999**	-0.0009	0.062**	-0.022**
X32	0.00093	0.00138**	-0.0195**	0.00191**	-0.0006	0.017**	0.001
Interaction							
X12	-0.00044	0.00032**	-0.0018**	0.00029	0.0007**	0.000	-0.002
X13	-0.00005	0.00042**	0.0116**	-0.00143**	-0.0003**	-0.010**	0.009**
X23	-0.00072	0.00170**	-0.0436**	0.00203	-0.0009**	0.040**	0.003
R2	59.0	98.2	99.5	90.8	82.3	99.5	90.6
R2adj.	40.6	97.4	99.2	86.7	74.4	99.3	86.4
Lack-of-fit	0.150	1.850	233.44	79.34	0.018	0.089	0.652
Model	*	*	*	*	*	*	*

$Y = \beta_o + \beta_1X_1 + \beta_2X_2 + \beta_3X_3 + \beta_{11}X^2{}_1 + \beta_{22}X^2{}_2 + \beta_{33}X^2{}_3 + \beta_{12}X_1X_2 + \beta_{13}X_1X_3 + \beta_{23}X_2X_3$; X_1 = Barrel temperature, X_2 = Feed Moisture content, X_3 = feed blend composition, CHO = Carbohydrate, * and ** = significant at 5% and 1% level of probability respectively

3.5. Numerical Process Optimization

MINITAB's Response Optimizer was adopted for simultaneous numerical optimization of the multiple responses, to search for a combination of independent variables levels that simultaneously satisfy the target requirement placed on each response and factors. Anuar *et al.*, (2013) and Gupta *et al* (2014) suggested that numerical optimization require that goals (None, Maximum, Minimum, Target or Range) should be set for the independent variables and response where all goals are combined into one desirable function. In this study, sets of conditions that will meet all the goals, the independent variables (i) barrel temperature (100-140°C), (ii) feed moisture content (15-25g.100g-1) and (iii) feed blend composition (8-24g.100g-1) were all set within range, while protein, water swelling capacity, minerals, fibre, amino acid (lysine) and high acceptability score were set at 5. Gupta et al. (2014) reported that the 'importance' score of a goal is within 1 to 5 and setting goal importance at 3 indicates that the variable is considered to be equally important, but Anuar *et al*. (2013) reported that when it is set at 5, the response target objective is to meet the objective of getting response at maximum level as applied in this study (Tables 7). The optimum independent variables were found to be 120°C barrel temperature, 24g/100g cowpea formulation and 20g/100g feed moisture composition which produce 99.02% dispersibility, 24.13g/100g protein, 73.44% carbohydrate and 388.82kcal/100g calorie, and lysine of 5.02mg/g protein. These results indicated that optimum values can be obtained in the extrusion of rice and soybean that can satisfy both nutritional and functional requirements of instant porridge consumers.

Table 6. *Estimated regression equation coefficients for response variables (amino acid profiles) in rice-cowpea extrudates.*

Term	Lysine	Isoleucine	Lucien	Valine	Methionine + Cystine	Threonine	Tryptophan + Phen.	Trypt.
Constant	-2.6686*	-4.5332*	10.5140*	11.9071*	2.3624	-4.4485	7.0172**	12.4167**
Linear								
X_1	0.0633**	0.0729	-0.1743*	-0.1714**	-0.0361*	0.0758	-0.0676**	-0.1781**
X_2	0.3289*	0.1826	0.2673	0.0460	0.0357	0.1337	0.0539	-0.0162
X_3	0.0329	0.1509	0.2458	0.0643*	0.0315	0.1371**	-0.0875**	0.0003
Quadratic								
X_1^2	-0.00005	-0.0002	0.0015*	0.0008*	0.0003*	-0.0002	0.0004**	0.0009**
X_2^2	-0.00325*	-0.0021	0.0103*	0.0019	0.0022*	-0.0015	-0.0008	0.0036
X_3^2	0.00014	-0.0010	0.0029*	-0.0012	0.0009*	-0.0004	0.0025**	0.0017*
Interaction								
X_{12}	-0.00151*	-0.0007	-0.0052*	-0.0010*	-0.0010*	-0.0008	-0.0004*	-0.0012*
X_{13}	-0.00084*	-0.0009	-0.0035*	-0.0001	-0.0004*	-0.0014*	-0.0007*	-0.0008*
X_{23}	0.00103*	-0.0007	0.0008	-0.0008	0.0006	0.0015	0.0040	0.0018
R^2	99.90	98.90	98.90	88.90	99.50	99.50	97.30	96.80
R^2_{adj}	99.70	97.00	97.00	79.00	98.50	99.80	92.40	91.00
Lack-of-fit	0.563	0.672	0.607	0.0740	0.0652	0.081	0.121	0.128
Model	*	*	*	*	*	*	*	*

$Y = \beta_o + \beta_1X_1 + \beta_2X_2 + \beta_3X_3 + \beta_{11}X^2{}_1 + \beta_{22}X^2{}_2 + \beta_{33}X^2{}_3 + \beta_{12}X_1X_2 + \beta_{13}X_1X_3 + \beta_{23}X_2X_3$; X_1 = Barrel temperature, X_2 = Feed Moisture content, X_3 = feed blend composition, * and ** = significant at 5% and 1% level of probability respectively.

Table 7. *Constraints and goals applied to derive optimum conditions of processing parameters and responses for rice-cowpea based formulations.*

Variables	Goal	Lower limit	Upper limit	Importance	Optimum level
Independent variables					
Moisture content (% w.b)	In range	8	24	3	20
Blend ration (% legume)	In range	15	25	3	24
Barrel Temperature (°C)	In range	100	140	3	120
Response variables					
Dispersibility (DSPLTY)	Maximize	98.29	99.23	5	99.02
Proximate composition					
Moisture content (%)	Minimize	0.08	0.92	3	0.63
Lipid content (%)	Minimize	0.04	0.61	3	0.41
Protein (%)	Maximize	19.00	29.06	5	24.13
Ash content (%)	Minimize	0.80	1.11	3	0.94
Fibre (%)	Maximize	1.62	2.88	5	2.00
Carbohydrate (%)	Minimize	68.15	77.21	3	73.44
Energy (kcal/100g)	Maximize	386.61	391.01	3	388.82
Amino acid composition (g/100g protein)					
Lysine	Maximize	4.12	6.00	5	5.02
Isoleucine	In range	2.41	3.20	3	3.00
Leucine	In range	6.45	10.74	3	8.14
Valine	In range	2.15	3.89	3	2.68
Methionine + Cysteine	In range	1.99	2.99	3	2.06
Threonine	In range	2.32	2.95	3	2.42
Tryptosin +Phenylalanine	In range	5.28	8.06	3	6.66
Tryptophan	In range	1.02	1.61	3	1.10

4. Conclusion

In this study, different optimum instant porridges were produce from several blends of broken rice fractions and cowpea. The cowpea was added to improve its nutritional quality. Response surface methodology and central composite design were adopted for the formulation and extrusion cooking. Statistically significant regression predictive models were fitted to demonstrate the relationship between the input and output variables. Optimum levels of the input variables that favour optimum production of porridge with high protein, calorie value and appreciable lysine level was achieved. This information can be adopted in the up-scale of extrusion cooking technology where rice and cowpea are the main ingredients. This will also sharpen the weaning food industry by providing wide opportunity for new food production using low grade rice and common legumes, thereby increasing the economic values of both local rice and cowpea.

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