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Development of an Optimized Cassava Starch-Based Custard Powder

Wasiu Awoyale[a,b], Lateef Oladimeji Sanni[c], Taofik Akinyemi Shittu[c], Abdulrazak Adesola Adebowale[c], and Mojisola Olanike Adegunwa[d]

[a]Postharvest Unit, International Institute of Tropical Agriculture, Ibadan, Oyo State, Nigeria; [b]Department of Food Science and Technology, Kwara State University, Malete, Kwara State, Nigeria; [c]Department of Food Science and Technology, Federal University of Agriculture, Abeokuta, Ogun State, Nigeria; [d]Department of Hospitality and Tourism, Federal University of Agriculture, Abeokuta, Ogun State, Nigeria

ABSTRACT

Custard powder is a dry-formulated food product primarily made from imported corn starch in developing countries. To reduce over-dependence on corn starch, this study investigated its replacement with yellow-fleshed cassava root starch (YfCRS) in custard powder formulation. Response surface methodology was used to develop an optimized cassava starch-based custard powder based on various combinations of YfCRS (90–98%) and whole egg powder (WEP) (2–10%). The result showed that the blends of YfCRS and WEP led to custard powder with wide functional, physicochemical, chemical, and micronutrient properties, with sensorily acceptable gruel. The second order response surface regression model accurately predicted most of these quality parameters ($R2 > 0.90$). The optimum formulation achieved to produce an acceptable custard was 90% YfCRS and 6.07% WEP. Therefore, this study showed that YfCRS might be a very good replacement for corn starch in the production of an acceptable custard.

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Introduction

Cassava contributes significantly to the nutrition and livelihood of up to 500 million people and thousands of processors and traders around the world, especially in the developing countries. Cassava is used as a raw material in the manufacture of processed food, animal feed, and industrial products. Wider utilization of cassava products can be a catalyst for rural industrial development and raise the incomes of producers, processors, and traders. It can also contribute to the food security status of its producing and consuming households (Plucknett, Phillips, & Kagho, 1998). Starch is one of the products from cassava. The end uses of cassava starch are potentially high due to its functionality in processed foods. The absence of the typical "cereal

CONTACT Wasiu Awoyale, wawoyale0101@gmail.com International Institute of Tropical Agriculture, PMB 5320 Ibadan, Oyo State, Nigeria.

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flavor" of corn and other cereal starches, the ability of higher swelling degree during cooking, and lower pasting temperature when compared with cereal starches are some of these properties. Additionally, the neutral flavor and white color of cassava starch are due to its low protein and fat contents (Wurzburg, 1989). These properties make cassava starch suitable for products like canned and powdered soups, instant desserts, sausages and processed meat, bakery products, and confectionery (Echebiri & Edaba, 2008). Nyerhovwo (2004) added that cassava starch could perform most of the functions where corn, rice, and wheat starch are currently used. Custard powder may be one of such possible product.

Custard powder is defined by Okoye, Nkwocha, and Agbo (2008) as a fine-textured dry food-product made from corn starch with the addition of salt, flavoring, and coloring agents and with or without the inclusion of egg yolk solids, vitamins, and minerals. As custard powder is known to be a carbohydrate-rich breakfast food consumed by many, its prolonged consumption might eventually lead to protein-energy malnutrition (PEM), which is common in most developing countries where cassava root is the major staple. Food insecurity or inadequate caloric intake is a major cause of death in the world, particularly in developing countries (Fiedler, 2009; Pelletier, Frongillo, Schroeder, & Habicht, 2005; Zimmermann & Hurrell, 2007). Chronic malnutrition, or insufficient intake of essential nutrients, affects more than two billion people worldwide, contributing to considerable illness, disability, and mortality (Caulfield, Richard, Rivera, Musgrove, & Black, 2006; Pelletier et al., 2005). Collectively, nutrition-related deficiencies are responsible for approximately 35% of global child deaths (1–2.5 million per year) and 11% of the total global disease burden (Pelletier et al., 2005). Food supplementation is the process of increasing in a food the level of specific nutrients previously identified as inadequate and their intake by the use of another food rich in that specific nutrient. This is usually done to prevent malnutrition in developing countries (FAO, 2002). The levels of food supplementation depend on the nutritional needs of the consumers and on both estimated consumption of the supplemented food as well as availability of the supplement and on the regulations in the country.

Thus, the supplementation of cassava starch with high-quality animal protein product, such as whole egg powder (WEP), in the production of custard powder could improve its protein quality and quantity. The egg is one of the most versatile and near-perfect foods in nature. It is rich in protein, amino acids, vitamins, and most mineral substances; the yolk and white components are all high in biological value and readily digested. The egg is known to supply the best proteins besides milk (Ihekoronye & Ngoddy, 1985; Vaclavik & Christain, 2008).

Response surface methodology (RSM) consists of a group of mathematical and statistical procedures that can be used to study the relationships between

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one or more responses (dependent variables) and factors (independent variables) (Murphy, Gilroy, Kerry, Buckley, & Kerry, 2003). The effectiveness of RSM in optimization of ingredient levels, formulations, and processing conditions in food technology from raw to final products such as baked cassava cake (Gana et al., 2007), sausage (Colmenero, Barreto, Mota, & Carballo, 1995; Murphy et al., 2003), snack food (Thakur & Saxena, 2000), puri (Vatsala, Dharmesh, & Haridas, 2001), biscuits dough (Gallagher, O'Brien, Scannell, & Arendt, 2003), and potato cubes (Varnalis, Brennan, Macdougall, & Gilmour, 2004), among others, have been documented by different researchers. At present, no scientific studies of the optimization of the basic formulation of CbCP have been reported.

Therefore, this study is aimed at developing an acceptable custard powder produced from the blends of YfCRS and WEP using response surface methodology, thus complementing the means of replacing imported corn starch with cassava starch in food formulations.

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Materials and methods

Production of yellow-fleshed cassava root starch and whole egg powder

The yellow-fleshed cassava root starch (YfCRS) was produced from TMS01/1368 using the traditional method of starch extraction described by Oyewole and Obieze (1995) with slight modification. The whole egg powder was also produced from quality whole eggs as reported by Awoyale, Sanni, Shittu, and Adegunwa (2015).

Experimental design to produce cassava starch-based custard powder

Response surface methodology of Design-Expert (Version 7.0) (Design-Expert, 2005) was used to determine the experimental design and the ingredients' combination levels for the cassava starch-based custard powder (CbCP) formulation. The two essential components incorporated in the custard were YfCRS (90–98%) and WEP (2–10%). Other ingredients added to the blends were vanilla flavor (3.5 g) and sodium chloride (1.5 g). The five coded levels of YfCRS: -α (88.34%), −1 (90%), 0 (94%), +1 (98%), +α (99.66%) and WEP: -α (0.34%), −1 (2%), 0 (6%), +1 (10%), +α (11.66%) were incorporated into the design and were analyzed in 13 runs with two blocks (Table 1). The central point of the design was repeated five times to calculate the reproducibility of the method. The effect of these two independent variables (WEP x_1 and YfCRS x_2) on the responses (Y) was modeled using the second-order polynomial response surface.

The equation derived using the RSM for the prediction of the response variables was:

Table 1. Central composite design of the yellow-fleshed cassava root starch and whole egg powder combinations for cassava starch-based custard powder formulation.

	Coded values		Actual values	
Trials	x_1	x_2	x_1	x_2
1	0	0	6.00	94.00
2	0	0	6.00	94.00
3	+1	−1	10.00	90.00
4	+1	+1	10.00	98.00
5	-α	0	0.34	94.00
6	0	0	6.00	94.00
7	−1	+1	2.00	98.00
8	0	+α	6.00	99.66
9	0	-α	6.00	88.34
10	−1	−1	2.00	90.00
11	0	0	6.00	94.00
12	0	0	6.00	94.00
13	+α	0	11.66	94.00

x_1 = Whole egg powder, x_2 = Yellow-fleshed cassava root starch

$$Y = \beta_0 + \Sigma\beta_1 x_1 + \Sigma\beta_2 x_2 + \Sigma\beta_{11} x^2{}_1 + \Sigma\beta_{22} x^2{}_2 + \Sigma\beta_{12} x_1 x_2 + \varepsilon$$

Where β_o is the value of the fixed response at the central point of the experiment that is the point (0, 0); β_1 and β_2 are the linear, β_{11} and β_{22} are quadratic, β_{12} are the interactions regression terms, and ε is the random error term.

Functional and physicochemical properties

Bulk density

This was determined using the method outlined in the AOAC (2000). The flour sample (7 g) was weighed into a 50 ml graduated measuring cylinder. The cylinder was tapped gently against the palm of the hand until a constant volume was obtained, and the bulk density (BD) calculated as shown:

$$BD = \frac{\text{Weight of sample}}{\text{Volume of sample after tapping}}$$

Water absorption capacity

The water absorption capacity (WAC) was determined using the method described by Beuchat (1977). A sample (1 g) each was mixed with 10 ml of distilled water and blended for 30 s. It was then allowed to stand for 30 min and centrifuged at 3500 rpm for 30 min at room temperature. The supernatant was decanted and weight of water absorbed by the flour was calculated and expressed as WAC.

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Swelling power

This was determined in accordance with the method described by Leach, McCovwen, and Schoch (1959) with modification for small samples. A sample of 0.1 g was weighed into a weighed test tube into which 10 ml of distilled water was added and heated in a water bath at a temperature of 60°C for 30 min. This was continually shaken within the heating period. At the end, the test-tube was centrifuge at 2,200 rpm for 15 min to facilitate the removal of the supernatant, which was then carefully decanted and the weight of the starch paste taken. The swelling power (SWP) was then calculated by:

$$\text{SWP} = \frac{\text{Weight of starch paste}}{\text{Weight of dry starch sample}}$$

Solubility index

The solubility index (SI) was evaluated by weighing l g of sample into a test tube with the addition of 20 ml of distilled water. This was subjected to heating in water bath at a temperature of 60°C for 30 min. At the end of heating, it was subjected to centrifugation at 2,500 rpm for 20 min, 10 ml of the supernatant was decanted and dried to constant weight, and the solubility expressed as the percent by weight of dissolved starch from a heated solution (Kainuma., Odat, & Cuzuki, 1967).

$$\text{SI} = \frac{\text{Weight of solubles}}{\text{Weight of sample}} \times 100$$

Dispersibility

The dispersibility of the samples was determined by the method describe by Kulkarni, Kulkarni, and Ingle (1991). Samples were weighed (10 g each) into 100 ml measuring cylinder and distilled water added to reach a volume of 100 ml. The set up was stirred vigorously and allowed to settle for 3 h. The volume of settled particles was recorded and subtracted from 100. The difference was then reported as percentage dispersibility.

Pasting properties

The pasting characteristics of the flour samples were determined using a Rapid Visco Analyzer (RVA) (Model RVA-4C, Newport Scientific, Warriewood, Australia) interfaced with a personal computer equipped with the Thermocline Software supplied by the same manufacturer, as reported by Newport Scientific (1998).

Amylose content

This was determined per the method described by Williams, Wu, Tsai, and Bates (1958). The calculation was done using this equation.

$$\%\text{Amylose of sample} = \frac{\%\text{amylose of standard} \times \text{Absorbance of sample}}{\text{Absorbance of standard}}$$

The %amylopectin = 100 – amylose content

Chemical composition

The crude protein was determined by the Kjeldahl method using Kjeltec™ model 2300-protein analyzer, as described in Foss Analytical Manual, AB. (2003). The moisture, ash, fat, and pH values were determined using standard methods as described by AOAC (2000). The starch content was determined as reported by Dubois, Gilles, Hamilton, Rebers, and Smith (1956). Hydrogen cyanide content was determined using the method reported by Essers, Bosveld, Van der Grift, and Voragen (1993).

Total carbohydrate content was calculated by difference as reported by Koua et al. (2012).

$$\text{Carbohydrate content} = 100 - (\%\text{Moisture} + \%\text{Ash} + \%\text{Fat} + \%\text{Proteins})$$

The total energy was calculated as reported by FAO (2003).

$$\text{Total energy} = [(\text{Protein} \times 4) + (\text{Carbohydrate} \times 4) + (\text{Fat} \times 9)]\text{J}$$

Iron and zinc content

The iron and zinc content of the samples were determined using the method described by Jones, Benton, and Vernon (1990). The samples were ashed at 550°C, after which their ash was dissolved in 5 ml water and 15 ml HNO_3/HCl (1:3). The minerals were then determined using Atomic Absorption Spectrophotometer (Buck 205 model; Back Scientific, USA).

Determination of β-carotene contents

The β-carotene contents of the samples were determined using High-Performance Liquid Chromatography (HPLC) (Agilent 1200 series, Perkin Elmer, USA) as reported by Carvalho et al. (2012)

Sensory evaluation of cassava-based custard gruel

The CbCP gruel was prepared by mixing 20 g of each of the samples with 100 ml of tap water in a small plastic bowl. Thereafter, 80 ml of boiling water was added to each of the suspended samples to produce hot gruel. After

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preparation, a teaspoonful of sucrose (9 g) was added to each of the samples to improve its taste. The samples of gruel produced were then served hot to the panelist (Okoye et al., 2008). A 9-point hedonic preference scale tests were used to test the acceptability of the gruel. Twelve trained panelists were selected from the staff and graduate students of International Institute of Tropical Agriculture (IITA), Ibadan, and screened with respect to their interest and ability to differentiate food sensory properties as described by Iwe (2002). Selection tests include odor, flavor, appearance, and color identification. The panelists were presented with coded samples to evaluate the effect of YfCRS and WEP blends on the CbCP gruel overall acceptability with the 9-point hedonic scale, where 9 corresponds to like extremely and 1 corresponds to dislike extremely.

Data analysis

All analyses were carried out in triplicates (except sensory evaluation) and subjected to analysis of variance (ANOVA) using Statistical Analysis System (SAS) package (version 9.1, SAS Institute, Inc., Cary, NC) (SAS, 2008). Means were separated using Fisher's protected least significant difference test. The combined analysis of variance was used to study the main and interactive effect of the independent variables on the responses. The optimization of the independent variables (YfCRS and WEP) in the formulation of the CbCP was done using Design-Expert (Version 7.0) (Design-Expert, 2005).

Results and discussions

The response surface methodology fitted models for the functionaland chemical properties of the cassava starch-based custardpowder, and the overall acceptability of the custard gruel

Table 2 revealed the means of all the properties of the actual experimental combinations of the YfCRS with the WEP. The amylose content ranged from 18.68 to 20.92%, dispersibility 83.45 to 86%, solubility index 0.99 to 3.47%, protein 0.73 to 6.39%, hydrogen cyanide 0.41 to 0.47 mg HCN/kg, starch 73.55 to 82.37%, total energy 159.66 to 166.98 J, Fe 22.19 to 29.39 mg/kg, Zn 2.47 to 4.25 mg/kg, trans β-carotene 0.1712 to 0.1894 μg/g, cis β-carotene 0.0956 to 0.1046 μg/g, and the overall acceptability of the gruel ranged from 6.30 to 7.20, which were the likeness range.

The fitting of the experimental data to the common second order polynomial equation for each response was done to establish predictive models for varying the YfCRS and WEP used in the formulations. The analysis of variance (ANOVA) was performed to evaluate the lack of fit (LoF) and the

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Table 2. Actual experimental combinations of yellow-fleshed cassava root starch with whole egg powder and response values.

Runs	WEP (X_1)	YfCRS (X_2)	Amylose (%)	Amylopectin (%)	Dispersibilty (%)	SI (%)	Protein (%)	HCN (mg HCN/kg	Starch (%)	Fat (%)	TE (J)	Iron (mg/kg)	Zinc (mg/kg)	β-carotene (µg/g)			Overall acceptability
														Total	Trans-	Cis-	
1	6.00	88.34	19.66	80.34	84.00	3.47	3.91	0.43	77.41	5.73	164.90	26.24	3.48	0.1792	0.0796	0.0996	6.30
2	2.00	98.00	20.58	79.42	84.00	0.99	1.61	0.45	81.02	4.28	159.66	23.30	2.75	0.1866	0.0833	0.1033	7.00
3	11.66	94.00	18.68	81.32	83.50	1.50	6.39	0.41	73.55	7.32	165.17	29.39	4.25	0.1712	0.0756	0.0956	6.60
4	2.00	90.00	20.54	79.46	84.00	2.91	1.70	0.45	80.87	4.31	160.41	23.41	2.78	0.1863	0.08315	0.1032	7.20
5	6.00	94.00	19.76	80.24	83.49	2.47	3.69	0.44	77.73	5.64	163.45	26.10	3.39	0.1799	0.07995	0.1000	6.80
6	6.00	94.00	19.71	80.29	83.45	2.41	3.72	0.46	77.75	5.63	163.44	25.95	3.42	0.1797	0.07985	0.0999	6.70
7	6.00	94.00	19.73	80.27	83.52	2.45	3.73	0.47	77.76	5.65	163.41	25.96	3.45	0.1796	0.0798	0.0998	6.60
8	10.00	98.00	19.05	80.95	83.50	2.97	5.42	0.42	75.02	6.72	166.98	28.19	3.97	0.1743	0.07715	0.0971	6.30
9	0.34	94.00	20.92	79.08	86.00	2.49	0.73	0.46	82.37	3.70	159.67	22.19	2.47	0.1894	0.0847	0.1046	7.10
10	6.00	99.66	19.80	80.20	84.00	2.96	3.55	0.43	77.98	5.50	161.98	25.78	3.35	0.1803	0.08015	0.1002	6.30
11	10.00	90.00	18.90	81.10	84.00	3.44	5.82	0.41	74.40	6.99	166.20	28.69	4.09	0.173	0.0765	0.0965	6.50
12	6.00	94.00	19.70	80.30	83.53	2.44	3.74	0.42	77.69	5.67	163.39	25.97	3.44	0.1798	0.0799	0.0999	6.50
13	6.00	94.00	19.74	80.26	83.50	2.46	3.71	0.43	77.71	5.61	163.40	25.99	3.41	0.1795	0.07975	0.0998	6.90

SI = Solubility index, HCN = Hydrogen cyanide, TE = Total energy, YfCRS-Yellow-fleshed cassava root starch, WEP-Whole egg powder
All the figures are means of triplicate values (except sensory acceptability)

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significance of the linear, quadratic, and interactive effects of the independent variables on the dependent variables. The LoF test is a measure of the failure of a model to represent data in the experimental domain at which points were not included in the regression and which must not be significant for the model to be significant (Varnalis et al., 2004). The coefficient of determination (R^2) is defined as the proportion of the variability in the response variables, which is accounted for by the regression analysis (Mclaren et al., 1977). When R^2 approaches unity, the better the empirical model fits the actual data. The smaller R^2 is, the less relevant the dependent variables in the model have in explaining the behavior of variation (Filmore, Kramer, & Gerald, 1976; Myers & Montgomery, 2002). It was suggested by Yadav, Vahia, Mahadevan, and Joglekar (2008) that for good fit model, R^2 should be at least 80%. The adequate precision value should be greater than four and the calculated F-value must be greater than the tabulated F-value for the model to be significant (Design-Expert, 2005).

Table 3 depicts the response values, regression coefficient, the coefficient of determination (R^2), F-value, Lack of Fit (LoF), and adequate precision values of the functional and physicochemical properties of the CbCP. The R^2 for the CbCP response, amylose, and amylopectin were more than 80%,

Table 3. Coefficient of second-order polynomial regression models and the analysis of variance results for the overall effect of process variables on the functional and physicochemical properties of cassava starch-based custard powder.

Coefficient	Dispersibility (%)	WAC (%)	SWP (%)	SI (%)	BD (%)	Amylose (%)
β_0	83.51	76.23	6.64	2.46	69.79	19.74
β_1	−0.50*	−1.70**	−0.2	0.14	NA	−0.79***
β_2	−0.063	−0.11	−0.68	−0.39	NA	0.048***
β_{11}	0.50*	NA	0.44	−0.24	NA	0.032**
β_{22}	0.12	NA	0.7	0.37	NA	−3.38E-03
β_{12}	−0.12	NA	0.76	0.36	NA	0.028*
R^2	0.6968	0.5823	0.5287	0.6029	NA	0.9995
Adeq Precis.	5.342	7.755	3.642	5.309	NA	174.493
F-Value	3.22	6.97	1.57	2.13	NA	2,826.92
LoF	S	S	S	S	S	NS
Total individual effect (F-value of ANOVA)						
WEP, X_1	8.46*	13.88**	0.24	0.48	NA	14,052.89***
YfCRS, X_2	0.13	0.06	2.73	3.75	NA	52.67***
Combined effect of all variables (F-value) at:						
Linear level	2.96	6.97*	1.25	1.58	0.48	1,956.21***
Quadratic level	3.62	1.54	1.59	2.39*	1.11	10.29**
Interactive level	0.16	0.2	1.51	1.24	0.21	2.76

*$p \leq 0.05$, **$p \leq 0.01$, ***$p \leq 0.001$, NS-not significant ($p > 0.05$), WAC = Water absorption capacity, SWP = Swelling power, SI = Solubility index, BD = Bulk density, β_0 = Regression coefficient for constant, β_1 = Linear regression coefficient for egg (X_1), β_2 = Linear regression coefficient for starch(X_2), β_{11} = Quadratic regression coefficient for egg, β_{22} = Quadratic regression coefficient for starch, β_{12} = Interactive regression coefficient for egg and starch, R^2 = Coefficient of determinant, LoF = Lack of Fit, Adj. = Adjusted, Pred. = Predicted, Adeq. Prec. = Adequate Precision

[a]F-Tabulated = 3.86, [2]F-Tabulated = 3.97, NA-Not available, YfCRS-Yellow-fleshed cassava root starch, WEP-Whole egg powder

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indicating significant models while that of dispersibility, water absorption capacity (WAC), swelling power (SWP), solubility index (SI), and bulk density (BD) were below 80%, indicating insignificant models. Considering other criteria and the subjective nature of dispersibility and SI, the models were considered significant. The calculated F-values for all the responses of the CbCP were more than the tabulated value (3.97–4.10) except for dispersibility, SWP, and SI, indicating insignificance of the model for these properties. The LoF tests of all the custard powder responses were significant except for amylose, indicating significant of the model for this property. All the responses had adequate precision > 4 except for SWP, indicating insignificant model (Table 3). The effect of the independent variables (YfCRS and WEP) on the selected responses reveals that at linear level, WEP had significant negative effect on dispersibility ($p \leq 0.05$) and amylose content ($p \leq 0.001$). The YfCRS, on the other hand, had significant positive effect on amylose content ($p \leq 0.001$). The linear effect of the YfCRS on dispersibility was negative but not significant ($p > 0.05$). At quadratic level, WEP had significant positive effect on dispersibility ($p \leq 0.05$) and amylose content ($p \leq 0.01$). The interaction of WEP and YfCRS had a significant positive effect on the amylose content (Table 3). The F-value from the analysis of variance for all the responses, representing their total individual and combined effect at the linear, quadratic, and interactive level, showed that YfCRS and WEP significantly ($p \leq 0.001$) affected the amylose content, while only WEP significantly affected dispersibility ($p \leq 0.05$). The combined effect of the independent variables at linear and quadratic level was significant ($p \leq 0.01$) for amylose content (Figure 1), while SI ($p \leq 0.05$) (Figure 2) was significant at the quadratic level (Table 3).

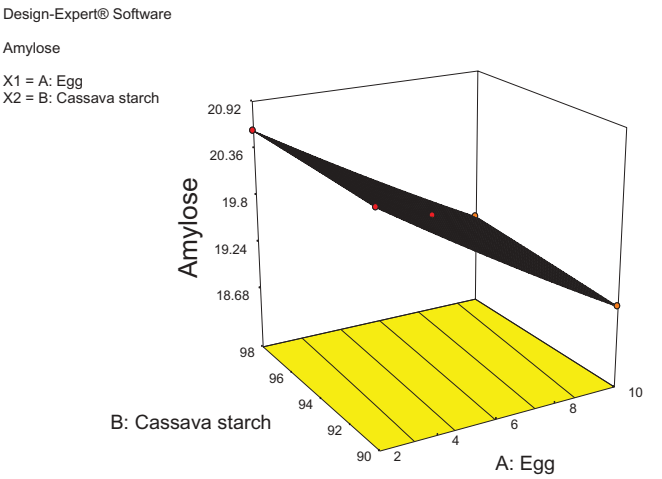


Figure 1. Response surface plots for the effect of whole egg powder and yellow-fleshed cassava root starch on the amylose contents of the cassava starch-based custard powder.

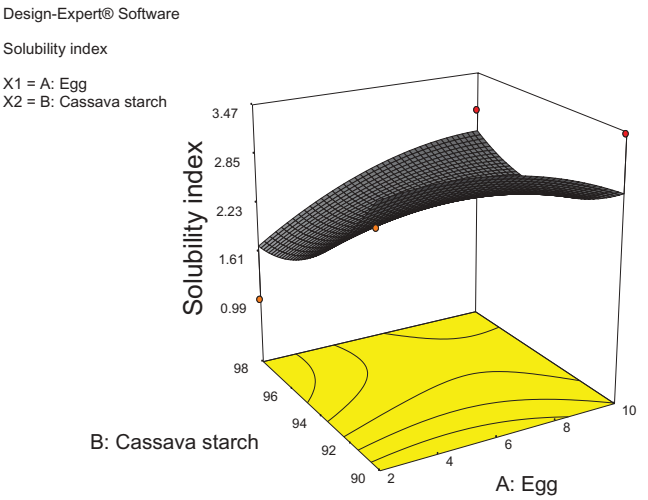


Figure 2. Response surface plots for the effect of whole egg powder and yellow-fleshed cassava root starch on the solubility index of the cassava starch-based custard powder.

Thus, the results of the functional and physicochemical properties of the CbCP showed that the model for amylose and amylopectin responses were highly adequate because they have satisfactory levels of R^2 of more than 80% with no significant LoF (Yadav et al., 2008). The negative effect of WEP on dispersibility and amylose content of the custard powder may be due to the high level of protein and fat as well as the low level of starch in its composition. The YfCRS, on the other hand, affected the amylose content of the custard powder positively, which could be due to the high level of starch containing amylose units. The model for the amylose and amylopectin content of the CbCP could explain about 99.95% of the variations in the amylose and amylopectin content. Thus, only 0.05% of the variation could be attributed to factors not included in the model. It was only the quadratic factor of the variables that significantly influenced the SI of the CbCP, and the model could explain about 60.29% of the variation, with 39.71% attributed to factors not included in the model.

The response surface methodology fitted models for the chemical properties of the cassava starch-based custard powder

The regression coefficient, R^2, F-value, lack of fit (LoF), and adequate precision values of the chemical properties of the CbCP are shown in Table 4. The R^2 for the responses; protein, ash, starch, fat, carbohydrate (CHO), and total energy (TE) were more than 80%, indicating significance of the model, while that of moisture content (MC), hydrogen cyanide (HCN), and pH, were below 80%, indicating insignificant models. The model for the HCN content was considered significant because of other criteria and the subjective nature

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Table 4. Coefficient of second-order polynomial regression models and the analysis of variance results for the overall effect of process variables on the chemical properties of cassava starch-based custard powder.

Coefficient	Protein[2] (%)	Ash[2] (%)	MC[2] (%)	pH[1]	HCN[2] (mg HCN/kg)	Starch[2] (%)	Fat[2] (%)	CHO[1] (%)	TE[1] (J)
β_0	3.71	1.95	8.53	4.75	0.44	77.72	5.64	80.19	163.24
β_1	1.99***	0.06	−0.088	0.078	−0.018**	−3.12***	1.28***	−3.24***	2.61***
β_2	−0.12***	5.52E-03	0.21	0.35	1.25E-03	0.20***	−0.078***	−0.014	−0.51
β_{11}	−0.078***	−0.25***	0.43	NA	NA	0.12***	−0.063***	NA	NA
β_{22}	7.13E-03	−0.16***	0.11	NA	NA	−0.015	0.010***	NA	NA
β_{12}	−0.078***	0.16**	−0.45	0.73	NA	0.12***	−0.06	NA	NA
R^2 (%)	0.9999	0.9424	0.6076	0.4401	0.6033	1.0000	0.9998	0.9676	0.8774
Adeq Precis.	405.255	11.921	4.148	5.826	8.097	639.488	295.76	35.955	17.283
F-Value	15,252.09	22.91	2.17	2.36	7.60	37,976.39	8,130.45	149.16	35.79
LoF	NS	S	S	S	NS	NS	NS	S	S
Total individual effect (F-value of ANOVA)									
WEP, X_1	75,799.26***	4.67	0.26	0.11	15.13**	3.960E+005***	40,372.50***	68.93***	298.32***
YfCRS, X_2	298.00***	0.04	1.44	2.18	0.076	1524.83***	150.54***	2.66	0.005
Combined effect of									
Linear level	2,235.76***	0.20	0.53	0.83	7.60**	2,420.47***	1,487.86***	35.79***	149.16***
Quadratic level	52.90***	46.22***	2.8	0.73	0.49	125.21***	42.41***	0.77	0.062
Interactive level	4.58*	1.57	2.53	4.79	0.14	4.69	4.35	0.72	3.20

*$p \leq 0.05$, **$p \leq 0.01$, ***$p \leq 0.001$, NS = not significant ($p > 0.05$), β_0 = Regression coefficient for constant, β_1 = Linear regression coefficient for egg (X_1), β_2 = Linear regression coefficient for starch(X_2), β_{11} = Quadratic regression coefficient for egg, β_{22} = Quadratic regression coefficient for starch, β_{12} = Interactive regression coefficient for egg and starch, R^2 = Coefficient of determinant, LoF =Lack of Fit, Adj. = Adjusted, Pred. = Predicted, Adeq. Prec. = Adequate Precision, [1]F-Tabulated = 3.86, [2]F-Tabulated = 3.97, NA = Not available, YfCRS = Yellow-fleshed cassava root starch, WEP = Whole egg powder

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of the response. The calculated F-values for all the responses were more than the tabulated value (3.97–4.10) except for MC and pH, indicating insignificance of the model for these properties (Table 4). The LoF test of all the responses was insignificant except for ash, MC, pH, CHO, and TE, indicating the inadequacy of the model for the properties. But, because of the subjective nature of the TE response, the model was considered significant (Table 4). All the responses had adequate precision > 4. Therefore, the responses protein, starch, fat, HCN, and TE contents of the CbCP were selected for developing the models. The effect of the independent variables on the selected responses revealed that at linear level, WEP had significant positive effect on the protein ($p \leq 0.001$), fat ($p \leq 0.001$), and TE ($p \leq 0.01$) contents but negative effect on HCN ($p \leq 0.01$) and starch ($p \leq 0.001$) contents of the CbCP (Table 4). The YfCRS, on the other hand, had significant ($p \leq 0.001$) negative effect on the protein and fat contents and with a significant ($p \leq 0.001$) positive effect on the starch content of the CbCP. The effect of YfCRS on the HCN was positive but negative for TE content ($p > 0.05$). At quadratic level, WEP had significant ($p \leq 0.001$) negative effect on protein and fat contents, and the effect was positive for starch content ($p \leq 0.001$) of the CbCP. The interaction of WEP and YfCRS had a significant ($p \leq 0.001$) negative effect on protein and fat content, while the effect was positive for starch content ($p \leq 0.001$) of the CbCP (Table 4). The F-value for all the CbCP responses, representing their total, individual, and combined effect at linear, quadratic, and interactive levels revealed that YfCRS and WEP significantly ($p \leq 0.001$) affected the protein, starch, and fat content of the CbCP, while the HCN ($p \leq 0.01$) and TE ($p \leq 0.001$) contents were significantly affected only by WEP (Table 4). The combined effect of the independent variables at both linear and quadratic level was significant ($p \leq 0.001$) for protein (Figure 3), starch (Figure 4), and fat contents

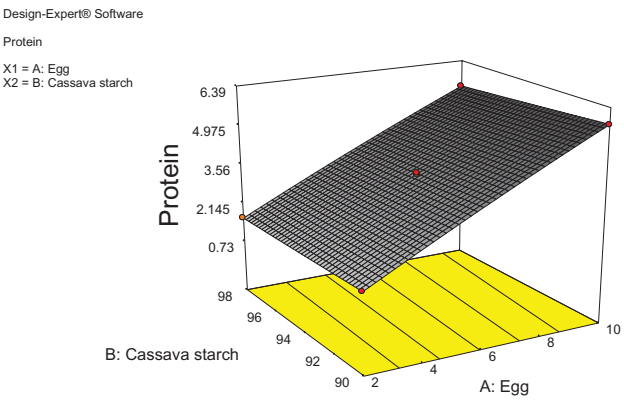


Figure 3. Response surface plots of the effect of whole egg powder and yellow-fleshed cassava root starch on the protein content of cassava starch-based custard powder.

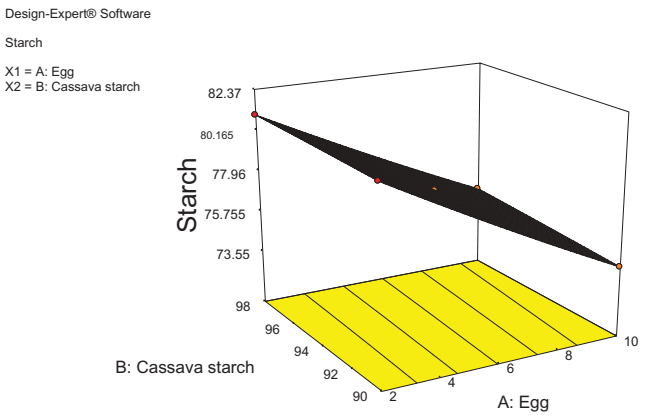


Figure 4. Response surface plots of the effect of whole egg powder and yellow-fleshed cassava root starch on the starch content of cassava starch-based custard powder.

(Figure 5), while that of HCN ($p \leq 0.01$), CHO ($p \leq 0.001$) and TE ($p \leq 0.001$) contents of the CbCP were significant at the linear level only. Protein content was the only dependent variable that was significant ($p \leq 0.05$) at the interactive level (Table 4).

Consequently, the models for all the chemical properties of the CbCP were highly adequate ($R^2 > 80\%$) excluding that of MC, HCN and pH value, due to the low level of R^2 ($R^2 < 80\%$) (Yadav et al., 2008). There was a significant influence of the linear and quadratic factors of YfCRS and WEP on the protein, starch, and fat content of the CbCP, while only the linear factor of these responses significantly influenced the HCN and TE of the custard powder. The significant positive effect of WEP on the protein, fat, and TE contents of the CbCP could be attributed to the high protein and fat content

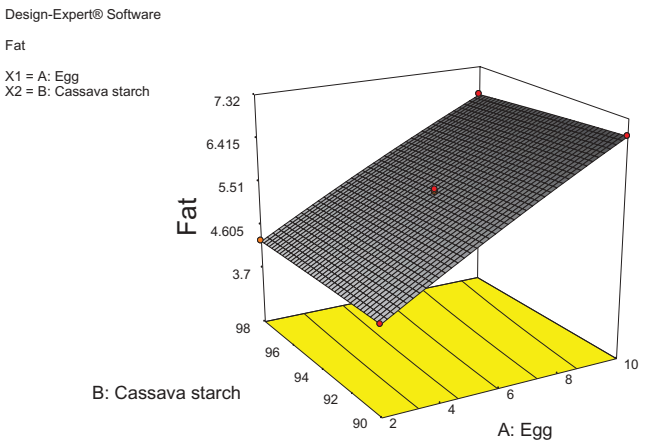


Figure 5. Response surface plots of the effect of whole egg powder and yellow-fleshed cassava root starch on the fat content of cassava starch-based custard powder.

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of the WEP, hence contributing to the TE of the product. Conversely, WEP has no HCN and starch, thus, the reason for its negative effect on these responses in the CbCP. The negative effect of the interaction between YfCRS and WEP on the protein and fat content and positive effect on the starch content of the CbCP could be because the bulk of the custard powder was from YfCRS and not WEP. In addition, the model could explain more than 87% of the variation in all the significant chemical responses excluding HCN (60.33%). Thus, only 13% of this variation could be attributed to factors not included in the model.

The response surface methodology fitted models for the micronutrient content of the cassava starch-based custard powder

The regression coefficient, R^2, F-value, LoF, and adequate precision values of the micronutrient contents of the CbCP are shown in Table 5. The R^2 for the CbCP micronutrient responses; iron, zinc, and the total, cis-, and trans- β-carotene contents were above 80%, indicating the significance of the model. The calculated F-values for all the responses were more than the tabulated value (3.97), the LoF test was insignificant and all the responses had adequate precision > 4, thus indicating the significance of the models for the CbCP. Therefore, all the CbCP micronutrient contents responses were selected for developing the models. At linear level, WEP had significant ($p \leq 0.001$) positive effect on the iron and zinc contents but the negative effect ($p \leq 0.001$) on total, cis-, and trans- β-carotene contents. The YfCRS, on the other hand, had significant negative effect on the iron ($p \leq 0.001$) and zinc ($p \leq 0.01$) contents but the positive effect ($p \leq 0.001$) on total, cis-, and trans β-carotene contents. At quadratic level, WEP had significant negative effect on iron ($p \leq 0.001$) and zinc ($p \leq 0.05$) contents, and the effect was positive for total ($p \leq 0.001$), cis- ($p \leq 0.01$), and trans- β-carotene ($p \leq 0.001$) contents. The interaction of YfCRS and WEP had a significant ($p \leq 0.01$) negative effect on iron content, while the effect was positive for total ($p \leq 0.05$), cis ($p \leq 0.01$), and trans β-carotene ($p \leq 0.01$) contents (Table 5). The F-values from the ANOVA for all the responses, representing their total, individual, and combined effect at the linear, quadratic, and interactive level, showed that YfCRS and WEP significantly ($p \leq 0.001$), affected all the micronutrient contents of the CbCP. The combined effect of the independent variables at linear ($p \leq 0.001$) and quadratic level was significant for iron ($p \leq 0.001$) (Figure 6), zinc ($p \leq 0.01$) (Figure 7), and the trans- (Figure 8) and cis- (Figure 9) β-carotene contents ($p \leq 0.05$) (Table 5).

The criteria for adequate models were met by all the micronutrient content of the CbCP as reported by Yadav et al. (2008). The observed significant positive effect of WEP on the iron and zinc content of the CbCP may be linked to the quantities of these minerals in the WEP compared to the

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Table 5. Coefficient of second-order polynomial regression models and the analysis of variance results for the overall effect of process variables on the micronutrient content of cassava-based custard powder.

Coefficient	Iron (ppm)	Zinc (ppm)	β-carotene (μg/g) Total	Cis-	Trans-
β_0	26.02	3.42	0.18	0.10	0.08
β_1	2.54***	0.63***	−6.417E-003***	−3.203E-003***	−3.209E-003***
β_2	−0.16***	−0.042**	3.945E-004***	1.936E-004***	1.972E-004***
β_{11}	−0.12***	−0.026*	4.063E-004***	1.313E-004**	1.531E-004***
β_{22}	−7.63E-03	2.00E-03	1.31E-04	3.13E-05	1.56E-05
β_{12}	−0.098**	−0.022	2.500E-004*	1.250E-004**	1.250E-004**
R^2 (%)	0.9996	0.9989	0.9992	0.9996	0.9997
Adeq Precis.	198.366	117.75	137.602	195.11	219.299
F-Value	3,655.61	1,288.16	1,762.03	3,534.67	4,468.2
LoF	NS	NS	NS	NS	Ns
Total individual effect (F-value of ANOVA)					
WEP, X_1	18,161.12***	6,399.22***	22,196.31***	17,569.87***	22,196.31***
YfCRS, X_2	69.67***	28.00**	83.86***	64.15***	83.86***
Combined effect of all variables (F-value) at:					
Linear level	1,680.42***	1,562.82***	1,642.21***	1,903.33***	1,642.21***
Quadratic level	16.95**	4.75*	22.00**	12.97**	22.00**
Interactive level	2.93	2.22	2.97	3.65	2.97

*$p \leq 0.05$, **$p \leq 0.01$, ***$p \leq 0.001$, NS = not significant ($p > 0.05$), β_0 = Regression coefficient for constant, β_1 = Linear regression coefficient for egg (x_1), β_2 = Linear regression coefficient for starch(X_2), β_{11} = Quadratic regression coefficient for egg, β_{22} = Quadratic regression coefficient for starch, β_{12} = Interactive regression coefficient for egg and starch, R^2 = Coefficient of determinant, LoF = Lack of Fit, Adj. = Adjusted, Pred. = Predicted, Adeq. Prec. = Adequate Precision, F-Tabulated = 3.97, YfCRS = Yellow-fleshed cassava root starch, WEP = Whole egg powder

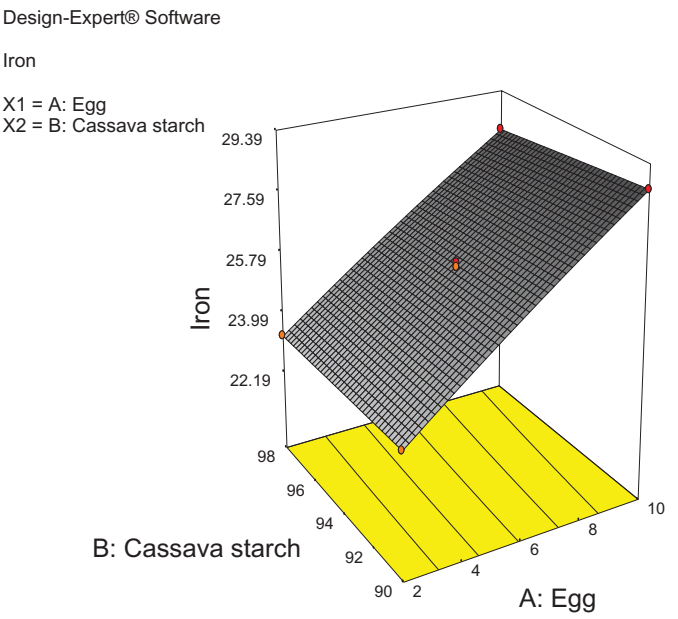


Figure 6. Response surface plots of the effect of whole egg powder and yellow-fleshed cassava root starch on the iron content of cassava-based custard powder.

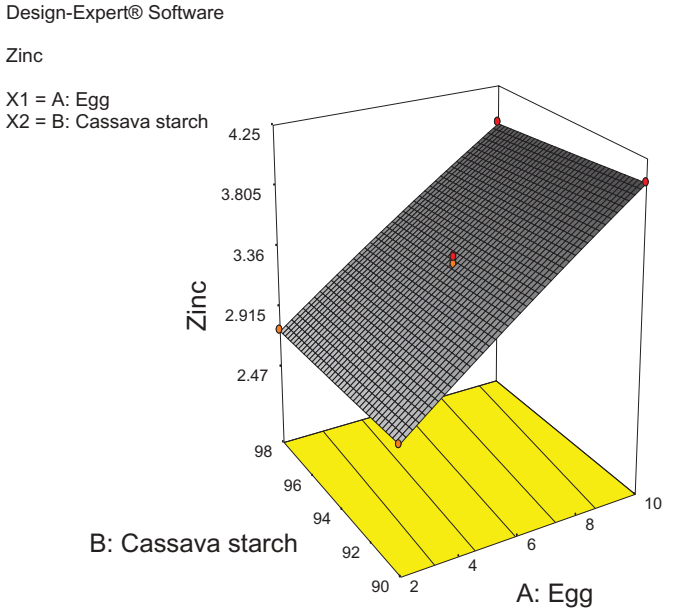


Figure 7. Response surface plots of the effect of whole egg powder and yellow-fleshed cassava root starch on the zinc content of cassava-based custard powder.

YfCRS. The WEP has little or no β-carotene, hence the reason for its considerable negative effect on the β-carotene contents of the CbCP.

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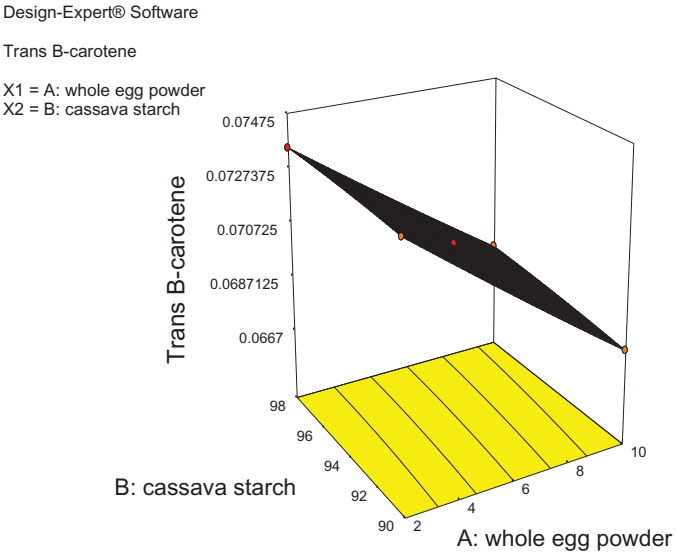


Figure 8. Response surface plots of the effect of whole egg powder and yellow-fleshed cassava root starch on the trans β-carotene content of cassava-based custard powder.

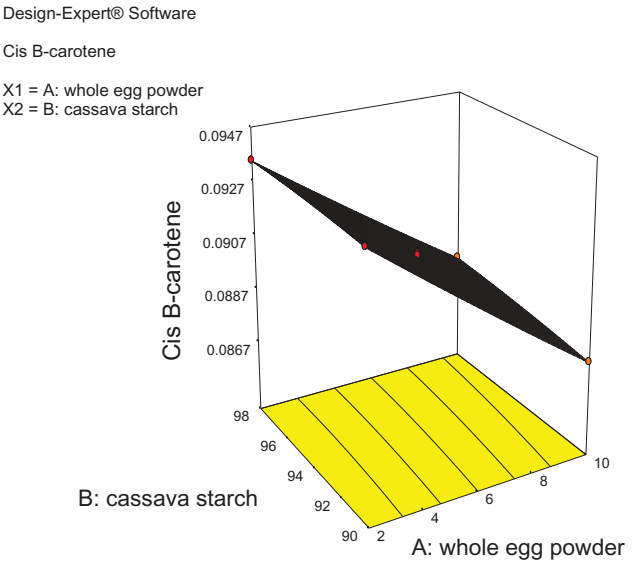


Figure 9. Response surface plots of the effect of whole egg powder and yellow-fleshed cassava root starch on the cis β-carotene content of cassava-based custard powder.

Contrary to the WEP, YfCRS has more of the β-carotene and little of the iron and zinc content, thus its significant positive effect on the pro-vitamin A content and negative effect on the iron and zinc content of the custard

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powder. This implied that the blends of YfCRS and WEP could contribute to the total micronutrient content of the CbCP in terms of its iron, zinc, and pro-vitamin A contents. Additionally, the considerable negative effect that the interaction of YfCRS and WEP had on the iron content of the CbCP could be because the bulk of the product was from YfCRS, which was low in iron compared to WEP. This was compensated for by the significant positive effect of these responses on the pro-vitamin A contents of the custard powder. More than 99.88% of these models could explain the variation in all the micronutrient content of the CbCP. So, only 0.12% of this variation could be attributed to factors not included in the model.

Optimization of the variables regarding the modeled functional, physicochemical, chemical and micronutrient content of the cassava starch-based custard powder

The modeled functional, physicochemical, chemical, and micronutrient contents as shown in Figures 3, 4 and 5 were useful for indicating the directions where the independent variables were to be changed to maximize the properties to produce the CbCP. These responses were used for the numerical optimization of the independent variables, and the criteria used are presented in Table 6. Consequently, in order to produce an acceptable CbCP, which may be highly soluble and disperse easily in water, might not weep easily after cooking and with high protein and energy contents, and also contribute to micronutrient contents, the response values of amylopectin, dispersibility, SI, protein, starch, fat, TE, as well as iron, zinc, and trans β-carotene content and the overall acceptability, were maximally optimized, while the amylose and HCN contents were minimized and the total and cis β-carotene contents were kept within range. The YfCRS and WEP were also kept within range as the independent variables while optimizing the dependent variables. By using these criteria, six desirable solutions were obtained, with one having the highest desirability (0.465513), which is 90% YfCRS: 6.07% WEP (Table 6). Thus, the consumption of the CbCP from the blends of 90% YfCRS and 6% WEP might contribute to the reduction of the incidence of malnutrition in developing countries.

Conclusion

The combination of yellow-fleshed cassava starch and whole egg powder lead to custard powder with wide functional, physicochemical, chemical, and micronutrients properties. The full quadratic model was found adequate to predict most of the properties of the cassava starch-based custard powder measured ($R^2 > 0.90$). The optimum formulation to produce an acceptable CbCP, which has minimum hydrogen cyanide and amylose contents with

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Table 6. Criteria for the optimization of the functional, physicochemical, chemical, and micronutrient content of cassava starch-based custard powder with the overall acceptability of the gruel.

					Solutions					
Constraints	Goal	Lower Limit	Upper Limit	Importance	1st	2nd	3rd	4th	5th	6th
WEP	is in a range	2.00	10.00	3.00	6.07	6.10	6.02	6.13	5.95	6.56
YfCRS	is in a range	90.00	98.00	3.00	90.00	90.00	90.00	90.00	90.00	90.00
Amylose	minimize	18.68	20.92	3.00	19.67	19.66	19.68	19.66	19.69	19.57
Amylopectin	maximize	79.08	81.32	3.00	80.33	80.34	80.32	80.35	80.31	80.43
Dispersibility	maximize	83.45	86.00	3.00	83.68	83.68	83.69	83.68	83.69	83.64
SI	maximize	0.99	3.47	3.00	3.21	3.21	3.21	3.21	3.22	3.18
Protein	maximize	0.73	6.39	3.00	3.88	3.90	3.86	3.91	3.82	4.14
HCN	minimize	0.41	0.47	3.00	0.44	0.44	0.44	0.44	0.44	0.43
Starch	maximize	73.55	82.37	3.00	77.46	77.43	77.50	77.41	77.55	77.06
Fat	maximize	3.70	7.32	3.00	5.73	5.74	5.71	5.75	5.69	5.90
TE	maximize	159.66	166.98	3.00	163.79	163.81	163.76	163.83	163.72	164.11
Iron	maximize	2.22	2.94	3.00	2.621	2.623	2.617	2.624	2.613	2.653
Zinc	maximize	0.25	0.43	3.00	0.347	0.348	0.347	0.348	0.346	0.355
Total β-carotene	is in a range	0.171	0.189	3.00	0.179	0.179	0.179	0.179	0.179	0.178
Trans β-carotene	maximize	0.076	0.085	3.00	0.080	0.080	0.080	0.080	0.080	0.079
Cis β-carotene	is in a range	0.096	0.105	3.00	0.100	0.100	0.100	0.100	0.100	0.099
Over acceptability	maximize	6.3	7.2	3.00	6.589	6.587	6.593	6.585	6.597	6.559
Desirability	-	-	-	-	0.465513	0.465509	0.465502	0.465498	0.46546	0.464708

YfCRS = Yellow-fleshed cassava root starch, WEP = Whole egg powder, SI = Solubility index, HCN = Hydrogen cyanide, TE = Total energy

maximum micro and macronutrients, solubility index, and overall acceptability was 90% YfCRS and 6.07% WEP. Therefore, this study showed that YfCRS might be a very good replacement for corn starch in the production of custard powder in developing countries.

Future research

In order to properly establish the replacement of corn starch with yellow-fleshed cassava root starch in the production of nutritionally rich custard powder, there is need for further supplementation with micronutrients such as vitamin A, iron, and zinc, among others, in order to meet recommended standard. Bioavailability of the supplemented micronutrients in the product also need to be investigated, as well as the consumer acceptability in developing countries.

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Disclosure statement

No potential conflict of interest was reported by the authors.

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