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Optimization of fermentation parameters for cereal-porridge production using Response Surface Methodology (RSM)

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Abstract

The possibility of the production of 'ogi' (a Nigerian cereal-porridge) with optimum quality was investigated using the Response Surface Methodology (RSM). Grains of white maize (*Zea mays)* and sorghum (*Sorghum bicolor*) were used separately and in combination (1:1 w/w), as the substrates for fermentations for a duration of 48h at ambient conditions. The starter cultures used in the controlled experiment consisted of 2 strains of *Lactobacillus fermentum* and a yeast, *Pichia kudriavzevii*. The organoleptic assessment of the 'ogi' showed that there was significant difference observed between the 'ogi' from the different substrates, with the maize-sorghum blend substrate being the most acceptable. Response surface optimization analysis using Box Behnken design revealed that the most desirable (0.89 desirability) conditions for 'ogi' production that could yield the following responses: pH of 5, protein content of 2.28% with overall acceptability of 8.5 were: temperature of 28 °C, substrate concentration of 50%, inoculum concentration of 4.0%, at 10h of fermentation. Response Surface Methodology was efficient in optimizing major fermentation conditions for the production of 'ogi' of high desirability from maize-sorghum blend, with the predictive and experimental values being closely related. The conditions could be employed in pilot study and industrialization of 'ogi' production.

Keywords: *Lactobacillus fermentum; Pichia kudriavzevii; Fermentation*; Optimization; 'Ogi'

1. Introduction

Fermentation has been reported to improve the nutritional value of their end products. It also provides food preservation, better sensory qualities of some foods, safer food products and the removal of anti-nutritional properties from some foods [1,2]. The contribution of fermented foods on our diet is of paramount importance in underdeveloped countries where a lot of fermented foods (especially cereal-based) are an integral part of daily food intake. Fermentation is indigenous to the Nigerian culture and is used to produce various foods. There are over 20 fermented foods and beverages in Nigeria, some are served as main meal while others are used as condiments. The production of these fermented foods depends on the traditional family settings [1, 2].

The cereal-porridge, 'ogi' is a major fermented food in Nigeria. It is produced by milling cereals such as maize, sorghum, or millet, sieving it wet and then allowing the starch to sediment and spontaneously ferment as a liquid substrate. The sediment is the' ogi' which is then boiled or stirred in hot water to form a gel-like porridge known as 'Ogi' porridge" [3,4]. 'Ogi' is an important weaning food for infants as well as a dietary staple breakfast or food for convalescent adults [5]. It is a fermented cereal product which is consumed across West African countries and known by different names such as Ogi among the Yorubas, Akamu among the Igbos/ Efiks/Ibibios and Akosa in Ghana [1, 5]. The traditional fermentation method used in 'ogi' production is a spontaneous process and microorganisms are not controlled [6].

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Food production on a large scale requires the optimization of the processing conditions without a reduction in the quality of the products. In order to make use of the generated data from important independent variables and employ statistical techniques to develop empirical models that are useful in predicting the optimal conditions for operations, response surface modeling is often used [7]. The production of 'ogi' is still a traditional or local household art in Nigeria. Commercial production is at a small scale despite the apparent increase in consumption due to ban on foreign foods and poor economic conditions in Nigeria. The industrial production of 'ogi' has been attempted only on a small scale since there is scanty information on a broad-based optimization process which is required for product development in industrial large scale production. This research therefore became necessary.

2. Material and methods

2.1. Processing of Maize and Sorghum into cereal-porridge ('ogi')

Healthy maize and sorghum grains were picked out from the chaff and separately steeped in sterile water (1:4 w/v) for 48 h to soften the kernel. A modified wet-milling and sieving laboratory method developed and reported by Ojokoh[3] was adopted for the processing of maize and sorghum grains into pastes and fermented. Three different types of samples were prepared as follows: Maize only (MZS), Sorghum only (SWM) and maize and sorghum blend (1:1, w/w) (MES). The samples were spontaneously fermented at ambient conditions for 48h. After the fermentation, 'ogi' samples were prepared for organoleptic testing.

2.2. Organoleptic Testing

This was done according to the method of Sherif *et al*. [8]. Clean potable water was heated and brought to the boil using a kettle. The supernatant of the fermented sample was then drained off and 500g of the thick sedimented paste was added to 150ml clean cold potable water in a clean, dried bowl. The mixture was homogenized by stirring gently with a clean, dried spoon until a smooth consistency was obtained. Then, while the heated water was still boiling, it was taken off the fire and poured into the homogenized sample in the bowl in a circular form while stirring, until the mixture became thick. The pouring of the hot water was then stopped and the stirring continued until a smooth, thick pap or 'ogi' was obtained. Ten cubes of white sugar (Dangote) were added and mixed thoroughly before sharing into 10 small labeled bowls and distributed to a ten-member semi-trained panel for sensory testing.

Quality characteristics of colour, aroma, taste and overall acceptability of the various 'ogi' samples were evaluated. Preference scores based on a nine-point hedonic scale was used, where 9 was scored as "like extremely" and 1 was scored as "dislike extremely". The results were analyzed statistically using analysis of variance (ANOVA) to determine differences and similarities

2.3. Controlled experiment

2.3.1. Sample preparation

The maize and sorghum grains were first washed and sterilized by autoclaving at 115°C for 15min and then cooled and processed (wet-milling and sieving).

2.3.2. Inoculum preparation and fermentation with starter culture

The method of Ali and Mustafa [9] was adopted for the inoculum preparation. The samples were then inoculated with 1ml each of the appropriate dilutions of the stock inoculum of the selected lactic acid bacteria (2 strains of *Lactobacillus fermentum*) and yeast (*Pichia kudriavzevii*) to give an approximate cell density of 1×10^9 cfu/ml and 1×10^7cfu/ml respectively. The fermentation was monitored to detect any possible microbial contaminants before the various analyses were carried out.

2.4. Analysis of Samples

2.4.1. Determination of Temperature, pH and Protein Content

The temperature was determined using a thermometer while the pH and protein concentration were determined according to the Method of Association of Official Analytical Chemists [10].

2.4.2. Determination of Substrate Concentration

The various substrate concentrations were determined by varying the ratios of the volumes of water to the weights of cereal grains during the wet-milling process.

2.5. Optimization of process parameters for 'ogi' production using Response Surface Methodology (RSM)

Box-Behnken design (BBD) of Response Surface Methodology (RSM) was employed in the optimization of the 'ogi' produced by controlled fermentation, using the consortium of two strains of *Lactobacillus fermentum* and a yeast (*Pichia kudriavzevii*) as starter cultures. The starter cultures were previously isolated from maize and surghum and identified by molecular method. The maize-sorghum blend sample (1:1 w/w) was used as the substrate since it was the most acceptable in the organoleptic test result. Independent variables included fermentation temperature, inoculum concentration, substrate concentration and fermentation time, while pH, protein content and overall acceptability of the 'ogi' were the responses.

A set of 27 runs of experiments was performed with three replicates at the centre points and 2 replicates for the factorial points. The ranges and the levels of variables used in the BBD are given in Table 1. Table 2 shows the experimental design matrix for optimization study. Each factor in the design was studied on three levels (- 1, 0, +1), with zero as the central coded value. These levels were derived from the results of the preliminary experiments. The interactions between the independent variables (temperature, inoculum concentration, substrate concentration, fermentation time) and the responses (pH, protein content and overall acceptability) were also determined during the optimization study. The design was carried out using Design-Expert version 11.0 (Stat-Ease Inc. Minneapolis, USA).

To test the estimation competence of the process, comparison between the actual responses and the predicted responses generated from RSM were drawn. Analysis of variance (ANOVA) and R^2 statistic aided evaluation of significant differences between various factors and the model's adequacy, which is best when close to 1. Lack-of-fit (a model's adequacy test tool), compares the pure error from measurement replications to the other lack of fit from the performance of the model.

F-value (the ratio of the lack-of-fit mean square to the pure error mean square) is the statistic parameter that determines the significance of the lack of-fit at a given significance level. Validation of the statistical model was based upon pH, protein content and overall acceptability estimation for 'ogi' production at Erlenmeyer flasks' level under the predicted conditions by the model. Sampling was carried out at desired intervals and pH, protein content and overall acceptability were determined. The Model was constructed using the equation below:

$$Y = \beta_0 + \sum\beta_i X_I + \sum\sum\beta_{ij} X_{IJ} + \sum\beta_{ii} x_i^2$$

Where: Y represents the predicted response, β_0 represents intercept term, β_i, β_{ij} and β_{ii} measure the variables effect χ_i, χ_{ij} and χ_i^2.

Table 1 Experimental ranges and levels of independent variables for Box-Behnken design (BBD) during the optimization of 'ogi' production

Independent variables	Range and levels		
	-1	0	+1
Temperature (°C), x1	26	28	30
Substrate concentration (%), x2	30	40	50
Inoculum concentration (%), x3	1	2.5	4
Fermentation time (h), x4	0	24	48

2.6. Data Analysis

The results obtained from the study were subjected to statistical analyses using SPSS 23. Analysis of Variance (ANOVA): Design Expert version 11 was used for the Response Surface Analyses (Box Behnken Design)

Table 2 Experimental design matrix for the optimization of 'ogi' produced using BDD

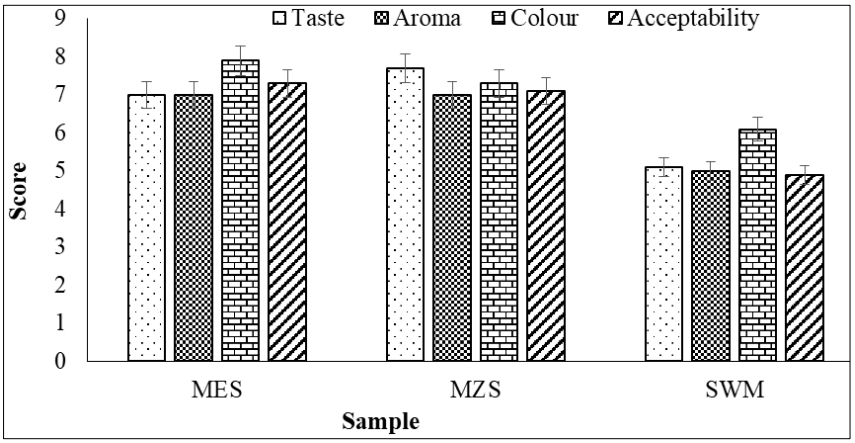
Run	A:Temperature (°C)	B: Substrate conc (%)	C: Inoculum conc (%)	D:Fermentation Time (h)
1	30	40	4	24
2	28	30	4	24
3	26	40	4	24
4	28	50	1	24
5	30	40	2.5	0
6	28	50	2.5	0
7	28	40	4	0
8	30	50	2.5	24
9	28	50	2.5	48
10	26	40	2.5	48
11	28	40	2.5	24
12	28	30	2.5	48
13	28	40	2.5	24
14	28	50	4	24
15	26	40	1	24
16	28	30	1	24
17	28	40	2.5	24
18	28	30	2.5	0
19	26	50	2.5	24
20	28	40	4	48
21	26	40	2.5	0
22	28	40	1	0
23	30	30	2.5	24
24	30	40	1	24
25	26	30	2.5	24
26	28	40	1	48
27	30	40	2.5	48

3. Results

3.1. Sensory quality of 'ogi' from maize, sorghum and maize-sorghum blend

The average preference scores of taste, aroma, colour and overall acceptability of 'ogi' produced from the different samples are shown in Figure 1. The following order of preference was observed for overall acceptability: MES(maize-sorghum blend) > MZS (maize) > SWM (Sorghum). Preference in terms of taste was highest in maize(7.7) and least in sorghum (5.1), while that of maize-sorghum blend was 7.0. In terms of aroma, maize and maize-sorghum blend were equally preferred (7.0) above sorghum 'ogi' (5.0). Preference scores of colour showed the order of maize-sorghum blend > maize >sorghum.

The analysis of variance (ANOVA) test of significance showed that the preference score for the 'ogi' made from maize-sorghum blend was highest and significantly different from samples made from maize only and sorghum only (P>0.5).



LEGEND: MZS=Maize; SWM=Sorghum; MES=Maize-Sorghum blend (1:1 w/w)

Figure 1 Average preference scores of taste, aroma, colour and acceptability of 'ogi' from different samples

3.2. Independent Variables and Responses

The composition of various experiments of the BBD for independent variables (temperature, substrate concentration, and inoculum concentration and fermentation time) and responses (pH, Protein (%), and overall acceptability) showed that the responses ranged from 3-5.6 for pH, 1.73-2.45% for protein and 5.2-8.9 for overall acceptability.

3.3. Fitting of model and ANOVA for the production of 'ogi' by a combination of two strains of *Lactobacillus fermentum* and *Pichia kudriavzevii* using maize-sorghum substrate

Table 3 Quadratic Model and lack-of-fit summary for the different responses (pH, protein and overall acceptability) involved in 'ogi' production

Responses	Parameters	Model	Lack of fit
pH	p-value	*0.0007	0.4194
	F-value	7.28	1.74
	Predicted coefficient of determination	0.4317	
	Adjusted coefficient of determination	0.7719	
	Coefficient of determination	0.8947	
Protein	p-value	*0.0056	0.3820
	F-value	4.65	1.98
	Predicted coefficient of determination	0.1535	
	Adjusted coefficient of determination	0.6627	
	Coefficient of determination	0.8443	
Overall Acceptability	p-value	*<0.0001	0.4598
	F-value	12.09	1.53
	Predicted coefficient of determination	0.6455	
	Adjusted coefficient of determination	0.8565	
	Coefficient of determination	0.9338	

*= P-value is significant

ANOVA and regression coefficients are listed in Table 3. For response A (pH), the Model F-value of 7.28 was obtained implying that the model is significant. This means that there is only a 0.07% chance that an F-value this large could occur due to noise. P-value of 0.0233 (< 0.0500) obtained indicates that the model terms are significant. The Lack of Fit F-value of 1.74 means the Lack of Fit is not significant relative to the pure error. There is a 41.94% chance that a Lack of Fit F-value this large could occur due to noise. Non-significant lack of fit is good as it means that the model can fit. Coefficient of determination (R^2) obtained from the model was 0.8947.

For the response, protein (%), Model F-value of 4.65 and P-value of 0.0056 (<0.0500) obtained indicate that the model terms are significant and there is only a 0.56% chance that an F-value this large could occur due to chance. The Lack of Fit F-value of 1.98 was obtained; there is only 38.20% chance that a Lack of Fit F-value this large could occur due to noise. The Lack of fit was not significant. The coefficient of determination from the model was 0.8443.

The response, overall acceptability had a Model F-value of 12.09 and P-value of <0.0001% (<0.0500) obtained indicate that the model terms are significant and there is only a 0.56% chance that an F-value this large could occur due to chance. The Lack of Fit F-value of 1.53 was obtained; there is only a 45.98% chance that a Lack of Fit F-value this large could occur due to noise. The Lack of fit was not significant. The coefficient of determination from the model was 0.9338.

3.4. Combined Effects of Substrate Concentration and fermentation temperature on pH, Protein Content and Overall Acceptability of "Ogi"

Figure 2 (a, b, c) shows the influences of substrate concentration and fermentation temperature on pH, protein content and overall acceptability of the 'ogi'. Substrate concentration of between 30% and 40% and temperature of between 26 ^{o}C-29 ^{o}C produced the lowest pH of 3.3 (acidic). On the other hand, Figure 2b shows the combined effects of substrate concentration and fermentation temperature on the protein content. The response surface of the parameter is similar to that of 2a, explaining the relationship between pH and concentration of metabolites, which include protein. According to the figure, substrate concentration of 30- 35% and 26^{o}C-30^{o}C of temperature produced protein of about 1.9%.

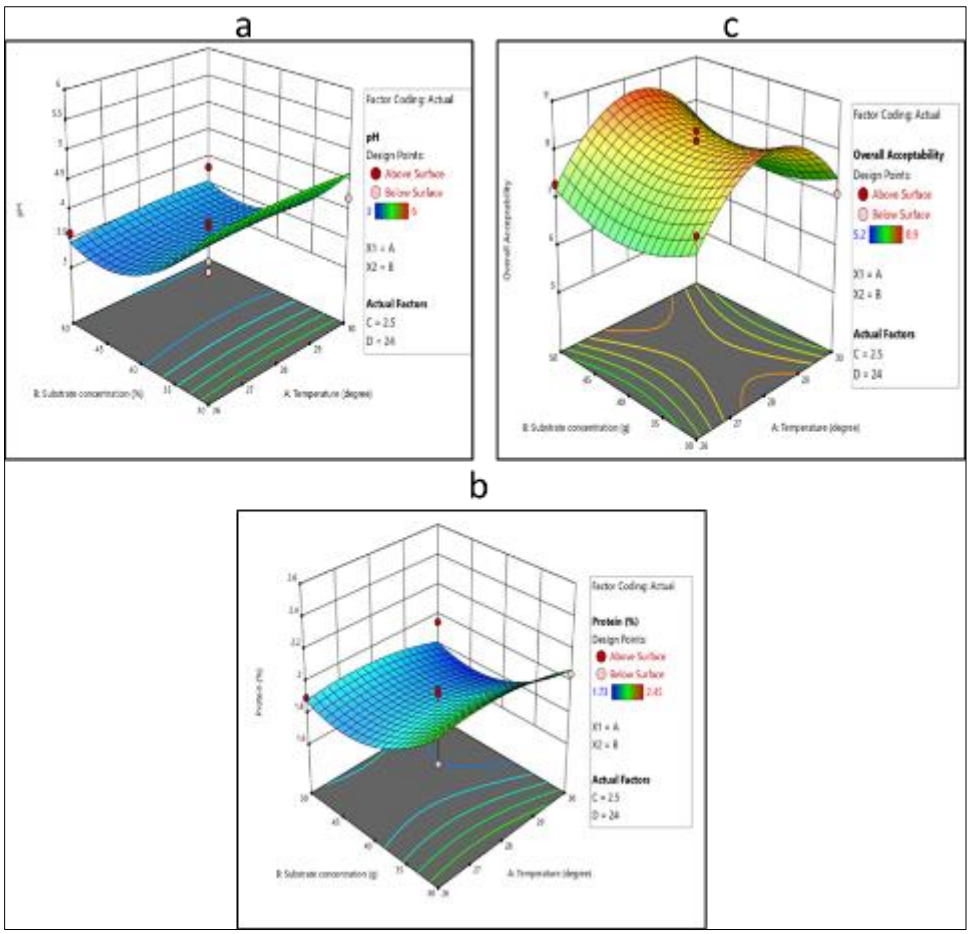


Figure 2 Response surface (3D plots) for the different responses (pH, protein and overall acceptability) involved in 'ogi' production as a function of temperature (°c) and substrate concentration (%)

The combined effects on overall acceptability as shown in Figure 2c produced a ridge response surface as the model indicating that the substrate concentration of between 35% -45% and temperature of 27°C -29°C scored the highest overall acceptability of 8.5. Changes (increases) in the variables therefore had a high influence on the overall acceptability.

3.5. Combined Effects of Fermentation Temperature and Inoculum Concentration on pH, Protein Content and Overall Acceptability of "Ogi"

The combined influence of fermentation temperature and inoculum concentration on the pH, protein content and overall acceptability of the "ogi" are shown on Figure 3 (a, b and c respectively). The response surface for the effects on pH showed a linear relationship, in which increases in the variables brought about only a slight decrease in pH from 3.8 to 3.7. That shows that increasing the inoculum concentration was not commensurate with the response. The highest pH of 3.8 was obtained by the combined influences of fermentation temperature range of 26 °C-28 °C and inoculation concentration range of 30%- 40%.

The influence of inoculum concentration and fermentation temperature on the protein content was also linear as increase in the variables also brought about an increase in the protein content. The inoculum concentration range and fermentation temperature range that had the maximum effects on protein (2.0%) contents were 3.1-4% and 26°C-29°C respectively.

On the other hand, the combined influence of fermentation temperature of 27°C-29°C and inoculum concentration of 2.2-3.4% brought about an overall acceptability score of 8.0, showing that the overall acceptability of 'ogi' does not require wide ranges of the variables.

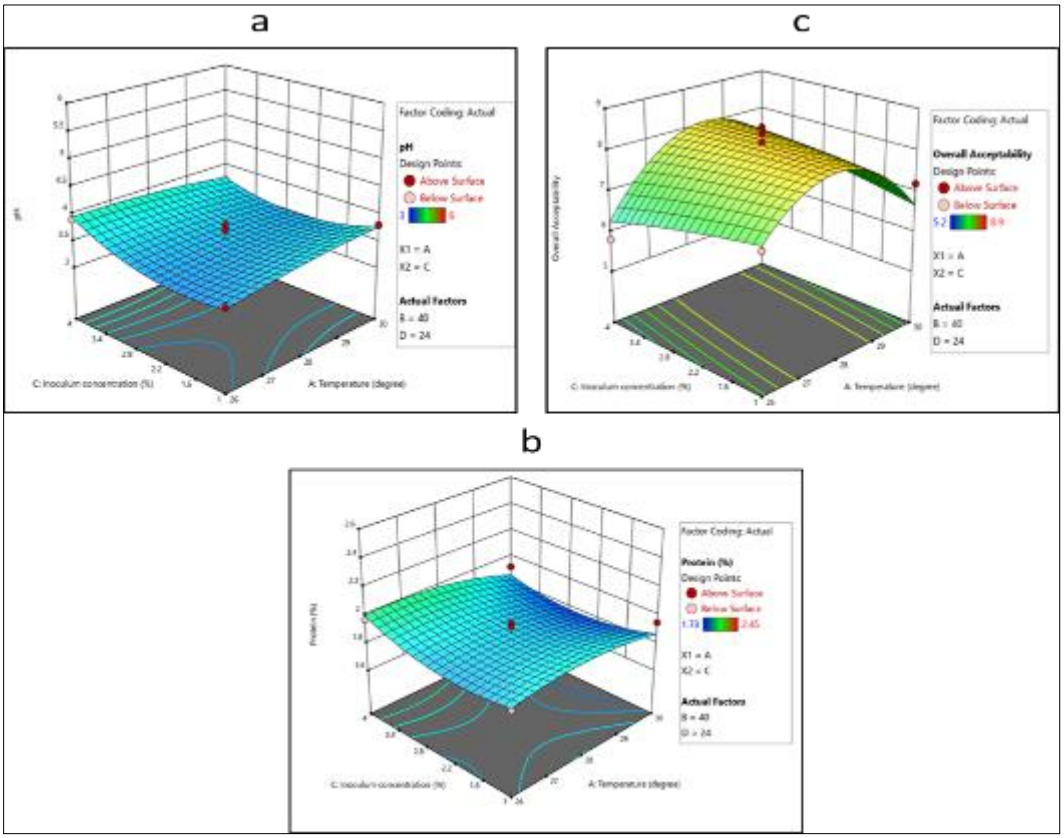


Figure 3 Response surface (3D plots) for the different responses (pH protein and overall acceptability) involved in 'ogi' production as a function of temperature (°C) and inoculum concentration (%)

3.6. Combined Effects of Fermentation Temperature and Fermentation Time on pH, Protein Content and Overall Acceptability

The combined influences of fermentation temperature and fermentation time on pH are shown in Figure 4a. The fermentation temperature range of 26-30°C and fermentation time range of 0-16h had the maximum influence on pH

changes from 5.5-3.0. The protein content (Figure 4b) value of 1.95% was achieved by the combined influences of the temperature range of 27-29^{o}C, fermentation time of between 0-16h. On the other hand, overall acceptability score (Fig 4c) of 7.5, being the maximum effect was influenced by a temperature range of 27^{o}C -29^{o}C at fermentation time of 12-36h. These results showed that fermentation temperature and time changes influenced pH and protein content less than they influenced the overall acceptability.

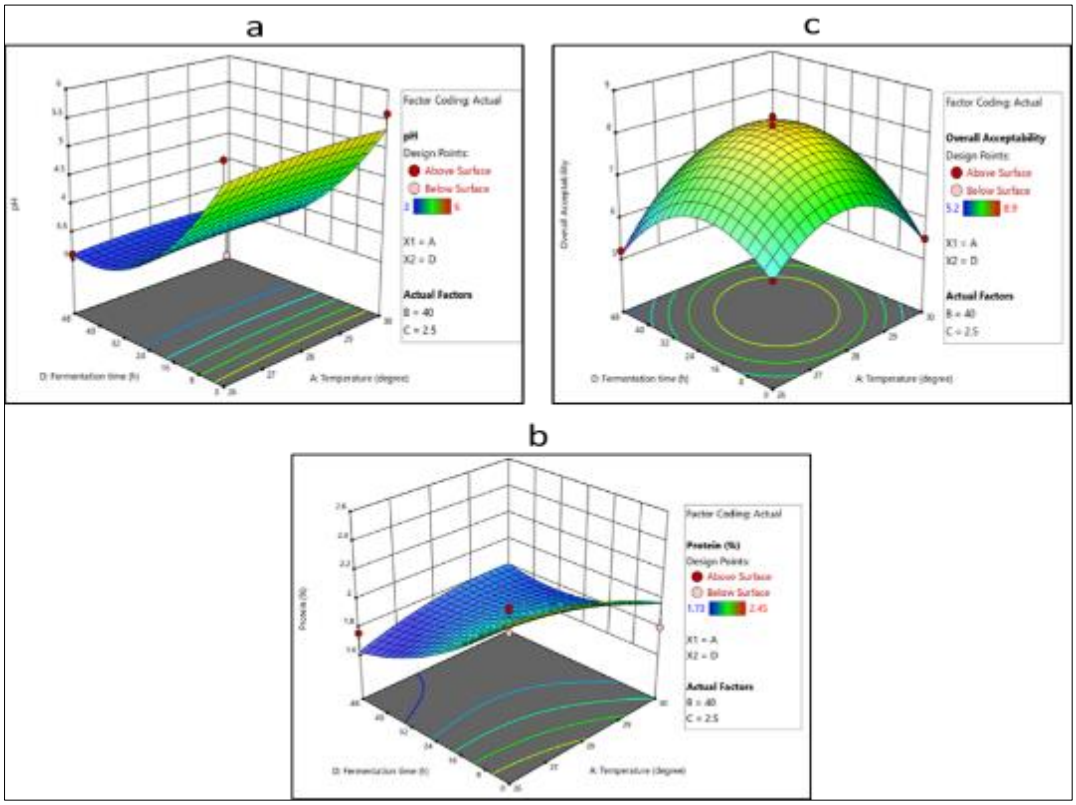


Figure 4 Response surface (3D plots) for the different responses (pH, protein and overall acceptability) involved in 'ogi' production as a function of temperature(°C) and fermentation time (h)

3.7. The Combined Effects of Substrate Concentration and Inoculum Concentration on pH, Protein Content and Overall Acceptability of the 'Ogi'

The effects are shown in Figure 5 (a, b and c). The response surface for the influences on pH (Figure 5a) showed that the inoculum concentration had a maximum effect at 2.2 – 4% while the maximum effect of substrate concentration was 30-35% for a pH of 5.3. The maximum effects on the protein content (Fig 5b) was produced with inoculum concentration of 1-4% and substrate concentration of 30-40%, whereas their maximum effects on overall acceptability, giving a high score of 8.9 were obtained at 1.0-3.4% and 40-50% ranges of inoculum and substrate concentrations respectively (Figure 5c). This indicated that the combined influences of the factors produced a highly acceptable "ogi".

3.8. Combined Effects of Substrate Concentration and Fermentation Time on pH, Protein Content and Overall Acceptability

The effects are shown in Figure 6(a, b and c). The maximum influences on pH 6.0 was due to the substrate concentration of 35-40% at temperature of 0-12h (Figure 6a), whereas maximum influences on protein content (2.4%) by the factors were within 0-16h of fermentation time at 40-45% substrate concentration (Figure 6b). A high overall acceptability of 8.5 was influenced by the action of 30-50% substrate concentration at 16-32h (Figure 6c). This indicated that to produce acceptable 'ogi' of good protein quality and low acidity, a high substrate concentration and fermentation time below 24h was required.

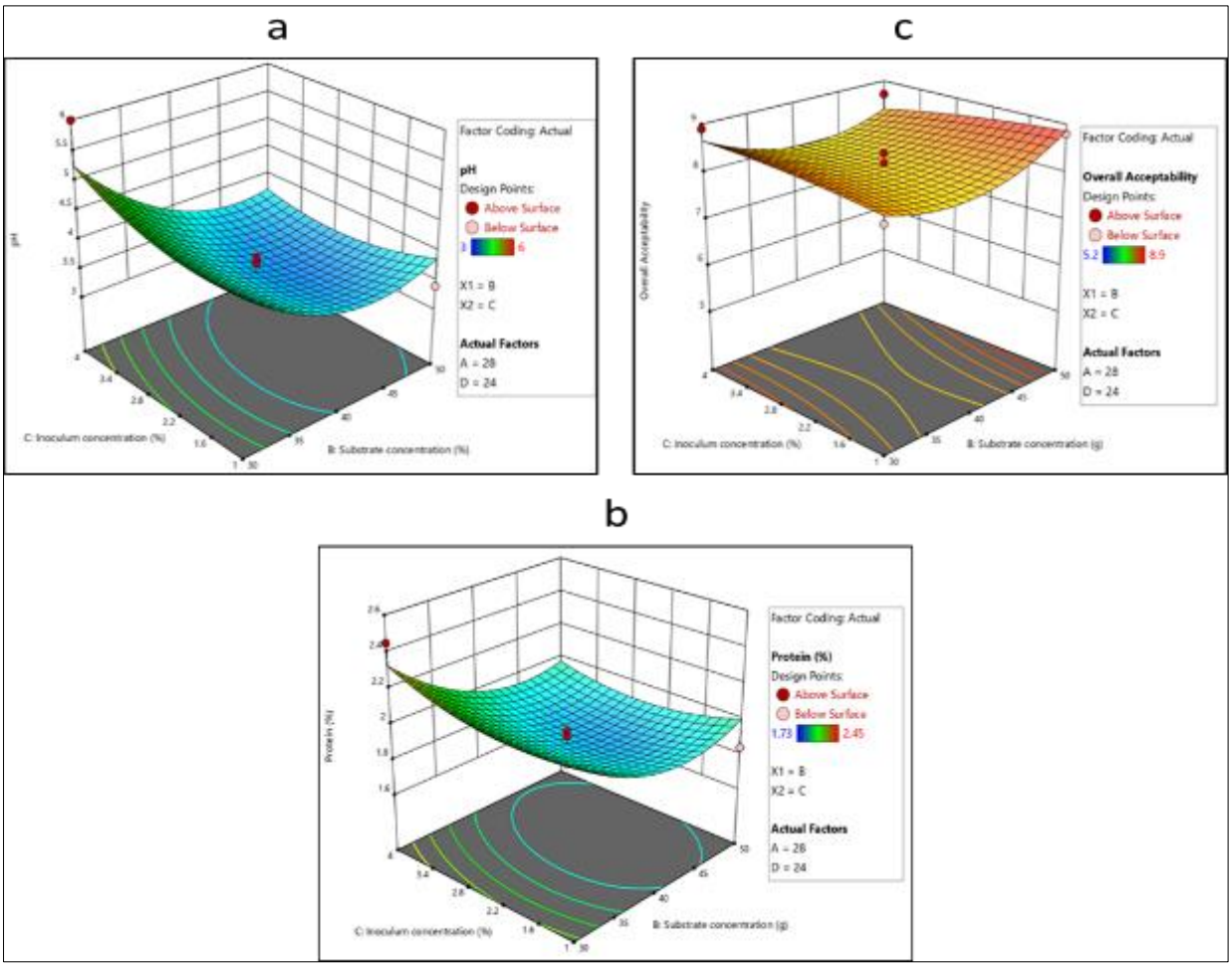


Figure 5 Response surface (3D plots) for the different responses (pH, protein and overall acceptability) involved in 'ogi' production as a function of substrate concentration (%) and inoculum concentration (%)

3.9. The Combined Effects of Inoculum Concentration and Fermentation Time on pH, Protein Content and Overall Acceptability of the 'Ogi'

The results are shown on Figure 7(a, b and c). The combined influences of inoculum concentration and fermentation time on pH of 5.5 were maximums at 2.2-4.0% at 0-8h respectively. The protein content of 2.3% was influenced by 2.2-4.0% inoculum concentration at a time range of 0-24h. The maximum combined effects on overall acceptability (8.0) were 1.0-4.0% inoculum at 16-32h. The results indicated that influence of inoculum concentration and time on the responses produced a less acceptable 'ogi'.

3.10. Analysis of Response Surface

The average values of the variables that had maximum effects on the various responses were as follows: Fermentation temperature of 27-29°C, substrate concentration of 30-35%, inoculum concentration of 3.4-4.0% and fermentation temperature of 1-10h.

3.11. Optimization Analysis

The optimization analysis showed that 28°C temperature, 50% substrate concentration, 4% inoculum concentration and 10h of fermentation produced 'ogi' of pH 5.0, protein concentration of 2.28% and 8.5 score of overall acceptability at 08.9 desirability.

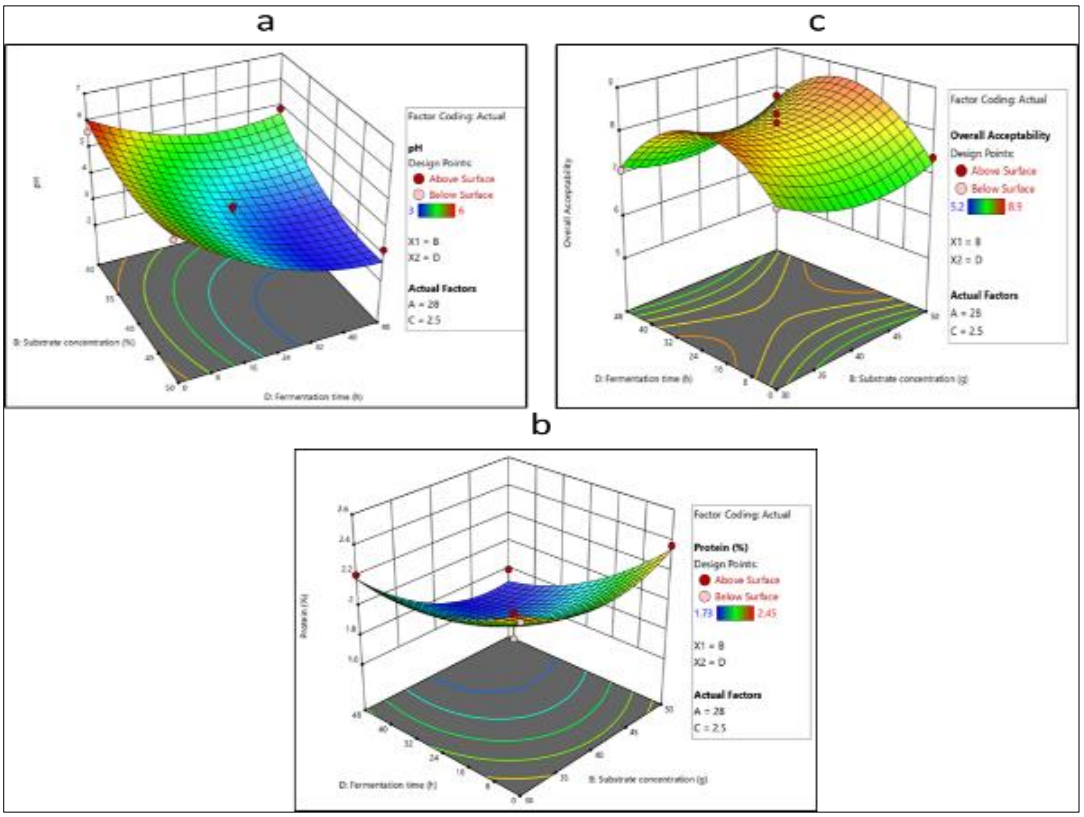


Figure 6 Response surface (3D plots) for the different responses (pH, protein and overall acceptability) involved in 'ogi' production as a function of substrate concentration (%) and fermentation time (h)

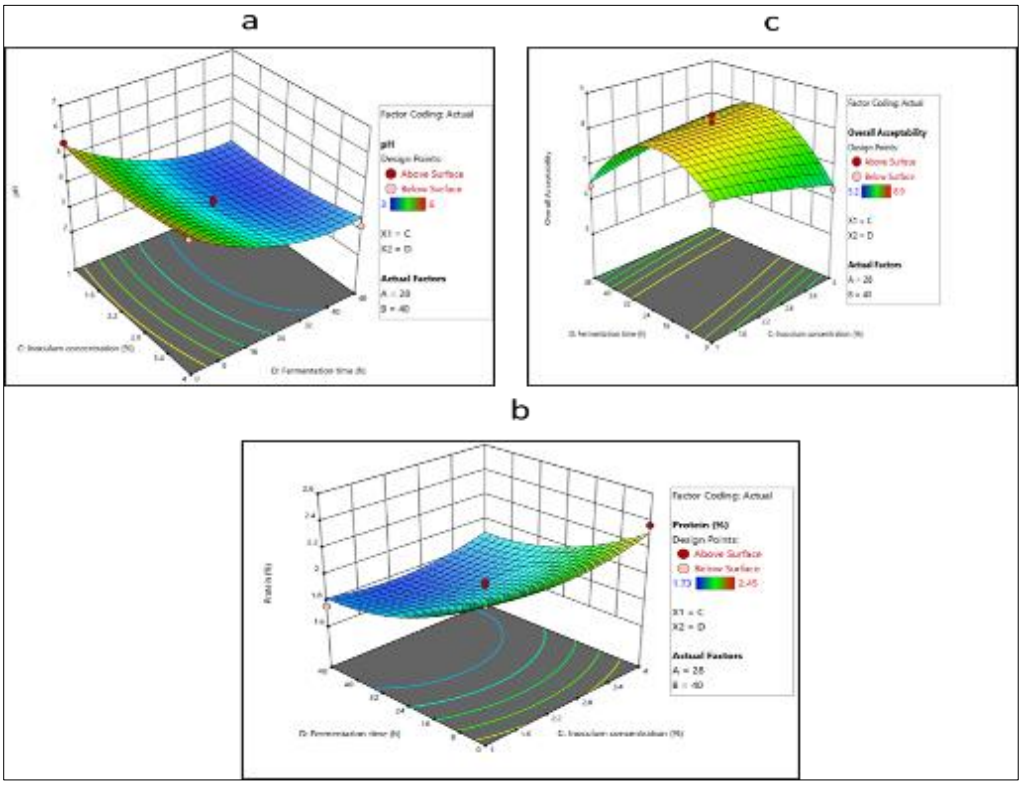


Figure 7 Response surface (3D plots) for the different responses (pH, protein and overall acceptability) involved in 'ogi' production as a function of inoculum concentration (**%**) and fermentation time (h)

4. Discussion

The optimization of some of the fermentation factors brought about 'ogi' of very high quality of 0.89 desirability. This was similar to the report of Bolaji *et al* [11] of which a desirable value was 0.99, in which soaking time and sedimentation time were the factors measured. Temperature had a maximum effect on pH, protein and overall acceptability at 27^0C to 29^0C. That was also similar to the report of Chinima *et al* [12] in which a temperature of 30^0C was achieved at 17h using 3% yeast concentration.

The results of this research have showed that the lactic acid bacterium, *Lactobacillus fermentum*, which doesn't require special supplement as nutrient can be used in combination with a probiotic yeast, *Pichia kudriavzevii* as starter cultures to produce 'ogi' of high quality (2.24% protein, a tolerable pH of 5 and acceptability of 85%) from the maize – sorghum blend, using the response surface methodology (RSM). The Box Behnken design of RSM which was used in the study was efficient and adequate since the predictive values and experimental values were related, and the model was able to predict the response value.

The preference scores of the sensory quality of the 'ogi' meals produced from the three different samples showed that the maize-Sorghum meal had the highest scores, and was significantly different from the other samples. The preference of the sample may be related to the factors that affect the sensory judgement of panelists. Such factors may be physiological, emotional, colour distraction and time of the day [8]. The colour of the maize -sorghum 'ogi' meal was more appealing and perhaps presented a distraction from other samples. The sorghum sample had the least scores in overall acceptability.

5. Conclusion

Response surface methodology (RSM) was efficient in optimizing the fermentation conditions for the production of 'ogi' with high protein content (2.24%), tolerable acidity level (pH 5) with high overall acceptability (85%). 'Ogi' produced by the combination of maize and sorghum substrate present statistically significant higher organoleptic qualities than those produced from either maize only or sorghum only. Substrate concentration and fermentation time were important factors in controlling the protein content and pH level of the 'ogi' produced in this study whereas fermentation temperature interaction with inoculum concentration or fermentation time significantly affected the overall acceptability of the 'ogi' produced.

Compliance with ethical standards

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Disclosure of conflict of interest

All authors declare that there is no conflict of interest.

References

[1] Ohenhen RE, Ikenebomeh MJ. Shelf Stability and Enzyme Activity Studies of Ogi: A Corn Meal Fermented Product. *Journal of American Science*. 2007; 3(1): 38-42.

[2] Osungbaro TO. Physical and nutritive properties of fermented cereal foods. African *Journal of Food Science*. 2009; 3: 2–27.

[3] Ojokoh AO. A comparative study on the effect of traditional and improved methods of fermentation in the production of Ogi food. *Oriental Journal of Chemistry*. 2009; 25(3): 471 – 476.

[4] Muller HG. Traditional cereal processing in Nigeria and Ghana. *Ghana Journal of Agricultural Science.* 1970; 3: 187 – 191.

[5] Adegbehingbe KT. Microbiological and nutrient studies of fermented cooked Lima bean seeds. *Global Journal Biology, Agriculture and Health Sciences*. 2014; 2: 94 – 101.

[6] Mbakwem – Aniebo C, Udemgba G. Microbiological quality of untreated and salt – treated Ogi (Akamu) kept at room temperature. *Nature and Science*. 2012; 10(8): 26 – 29.

[7] Box GEP, Draper N. Response surfaces, mixtures and ridge analyses (Second ed.). Hoboken: John Wiley &Sons. 2007.

[8] Sharif MK, Butt MS, Sheriff HR, Nasir M. Sensory evaluation and consumer Acceptability; in book: Handbook of Food Science and Technology. 2017; 362 -386.

[9] Association of Official Analytical Chemists (AOAC). Offical Methods of Analysis(18th) Edition. AOAC, Washington D. C., 2005.

[10] Ali AA, Mustapha MM. Use of starter Cultures of Lactic Acid Bacteria and Yeasts in the preparation of Kisra, a Sudanese Fermented food. Pakistan Journal of Nutrition. 2009; 8: 1349-1353.

[11] Bolaji OT, Adenuga-Ogunji L, Abengunde TA. Optimization of processing conditions of 'ogi' produced from maize using response surface methodology (RSM). Cogent food and Agriculture.2017; 3:1-10.

[12] Chinma CE, Ilowefah M, Muhammad K. Optimization of Rice Bran fermentation Conditions Enhanced by Baker's Yeast for Extraction of Protein Concentrate. Nigerian Food Journal. 2014; 32(1):126-132.