IPS Journal of Nutrition and Food Science
IPS J Nutr Food Sci, 3(2): 145-153 (2024)
DOI: https://doi.org/10.54117/ijnfs.v3i2.45

Determination of Viscosity, Syneresis and Acceptability of Optimized Yoghurt Analogues from African Yambean (*Sphenostylis stenocarpa Hams*) and Malted Red Rice (*Oryza glabberima*)

Blessing Chisomuaga Okoronkwo, Lucretia I. Barber and Patience C. Obinna-Echem

Department of Food Science and Technology, Rivers State University, Nkpolu-Oroworukwo, P.M.B 5080, Port Harcourt, Rivers State.
**Corresponding author e-mail:* blessedbbken@gmail.com

Abstract

Yoghurt analogues were produced using African yambean (*Sphenostylis stenocarpa Hams*) and malted red rice (*Oryza glabberima*). Optimization of the mixture components and process factor was carried out using combined optimal mixture design, viscosity, syneresis and acceptability as responses were analysed. The fitted regression model for viscosity, syneresis and acceptability were all significant (P<0.05) with coefficient of determination (R^2) values of 0.9992, 0.9983 and 0.9904 % respectively. The coefficient of variation (cv) values were 0.992, 6.58 and 6.58 % respectively. The lack-of-fit for the models were not significant ($P < 0.05$). The suitable mathematical models developed for the optimization of the fermentation variables in African yambean and malted red rice yoghurt is highly recommended for the development of a novel non-dairy probiotic yoghurt.

Keywords: Viscosity, Syneresis, Yoghurt analogue, African Yambean, Malted Red Rice

Article History
Received: 03 Mar 2024
Accepted: 15 Mar 2024
Published: 21 Mar 2024

Scan QR code to view•

How to cite this paper: Okoronkwo, B. C., Barber, L. I., & Obinna-Echem, P. C. (2024). Determination of Viscosity, Syneresis and Acceptability of Optimized Yoghurt Analogues from African Yambean (Sphenostylis stenocarpa Hams) and Malted Red Rice (Oryza glabberima). *IPS Journal of Nutrition and Food Science*, *3*(2), 145–153. https://doi.org/10.54117/ijnfs.v3i2.45

Introduction

Some factors that affect wellbeing includes lifestyles, increasing cost of health care among others (Barber *et al.*, 2021). Diet plays a major role in preventing diseases and promoting health, (Soomro *et al.*, 2002). There are scientific evidences that suggests that consumption of fermented foods, especially fermented milk products is associated with improved health outcomes (Dennise and Hutkins, 2020). Fermented milk products are good sources of protein, calcium, phosphorus, vitamins and carbohydrates (Daniella *et al.*, 2016). Moreso, fermented milk products containing probiotic organisms are regarded as healthier foods (Barber *et al.*, 2021). Non-dairy traditional beverages mainly based on cereals have long existed all over the world. Notwithstanding several new non – dairy probiotic beverages have been recently developed like oat yoghurt, soy-yoghurt and tigernut beverages (Nionelli *et al.*, 2014). African yam bean (*Sphenostylis stenocarpa)* is an underutilized legume. It is an important food substitute of cowpea in many parts of south eastern Nigeria where it is largely grown (Ukom *et al.*, 2014). The seeds have been preferred to other legumes in the past because they are filling, though cowpea is now the preferred legume due to its commercialization (Mbaeyi, 2011).

Nutritionally, the seed is rich in protein with values ranging between 19 and 30% (Klu *et al.*, 2000; Nwosu, 2013; Abiroye *et al.*, 2015; Adeomowaye *et al.*, 2015; Duodu and Apea-Bah, 2017; Anya and Ozung, 2019). The protein in AYB compares favourably with those in pigeon pea, chickpea, bambara and cowpea, the bean is also rich in dietary fibre (Ndidi *et al.*, 2014; Baiyeri *et al.*, 2018; Anya and Ozung, 2019; Nwosu, 2013., 2014; Ajiobola and Olapade, 2016) and important minerals such as calcium, iron, zinc, magnesium amongst others with values higher or comparable to soy and common beans (Abioye and Omotosho, 2015). The levels of sodium and copper are low (Duodu and Apea-Bah, 2017). The essential amino acid proportion in the protein of AYB is over 32% with lysine and leucine being predominant (Toyosi *et al.*, 2020). African yambean yoghurt –like product was developed by Taiwo & Zulfah (2014), fermented African yambean flour was produced by Okeke and Chikwendu (2015). Edith *et al.* (2018) also developed a yoghurt-like product from *Bambara* nut, Soybean and *Moringa oleifera.*

Rice (*Oryza sativa*) is a major cereal crop consumed as a staple food by over half of the world's population, its consumption is very high in developing countries (Cokro and Romulo, 2012). The lysine content of rice protein is between

3.5 and 4.0% making it highest among cereal proteins (Rathna, 2019). Rice with a red bran layer are called red rice (*Oryza glabberima*. The bran layer contains polyphenols and anthocyanin, and possesses antioxidant properties (Rood, 2000). Rice milk is an important food source considered worldwide due to its high antioxidant activity (Sangkitikomon *et al*., 2008). . Probiotic yoghurt analogue was developed by Uzuner *et al*. (2016) using rice milk, the author also reported the protein content of rice milk to be 2.53 and 3.43%, percentage fat to be 3.25-2.90% respectively.

Among diverse fermented milk products, yoghurt is most popular and more acceptable throughout the world (Kumar *et al*, 2015) because of its general positive image among consumers (Rood, 2000). Yoghurt is mainly produced from cow's milk. It may also be produced from fruits, cereals, legumes and non-ruminant animals etc (Erdogan and Zekai 2003).

Yoghurt possesses high nutritional value, some of the nutrients include calcium, zinc and vitamin B (El-kholy *et al*., 2011) and it also exerts bioactive effects. Some studies have also reported the production of plant based yoghurt from some legumes like soybeans, Bambara groundnut and African yambean (Zanhi and Jideani, 2012, Kolawole *et al*., 2015).

There are no reports on the production of yoghurt using malted red rice. Previous studies have focused on the production of probiotic and fermented dairy products made using vegetable, cereals and legume based raw materials, however there is a limited number of studies on the usage of malted red rice milk and African yambean in the production of yoghurt analogue.

Materials and Methods

African Yambean (*Sphenostylis stenocarpa Hams*)

The seeds of the African yam bean (*Sphenostylis stenocarpa Hams*), cream coloured variety (Odudu) was purchased from retailers at Umuahia Main Market, Abia State, Nigeria.

Red rice (*Oryza glaberrima*)

The red rice was sourced from Tovia Farms Ogun state, Nigeria.

Ingredients

The stabilizer used was purchased from De Giant Bakers Port Harcourt, Rivers State.

Chemicals

The chemicals were of analytical grade and were obtained from Joechem Chemicals Choba Port Harcourt, Rivers State. These include sodium bicarbonate, mineral oil. Sodium hydroxide, crystal violet, lugol iodine solution, safranin, 9and 97% ethanol hydrogen peroxide, magnesium chloride, kjedal catalyst tablet 0.30% (w/v) oxgall-bile, 1.5% agarose gel, hydrogen chloride, phosphate buffer saline and concentrated sulphuric acid (H_2SO_4).

Malting of red rice

The rice was malted by adoption of barley malting protocols according to Kunze (2005), Steeping was done by submerging 200g of sorted undihulled red rice in 1000ml of portable water at 25ºC for 36h. The steep cycle involved alternating 12h wet-steep with 45m air-rest period. After steeping, the grains were couched (heaped) on jute bags previously sterilized with dry heat. The grains were germinated within at the temperature of 25ºC and were removed after the second day of germination. Kilning was done in a hot air oven at 70ºC for 3h. Kilned grains were manually de-rooted by rubbing off with hand and winnowed to remove the rootlets and dust.

Production of malted red rice milk

Hundred grams (100g) of the dehulled malted red rice was dry milled in a blender. Afterwards it was sieved to obtain the powder. The powder was then mixed with water in the proportion of 1:10 w/v to form the slurry. The slurry was heated in a water bath at 100ºC for 30min. The heated liquid was poured into sterile glass container, cooled to 25ºC and then stored at 4ºC in the refrigerator for fermentation (Cokro and Romulo 2012).

Production of African yam bean milk

The AYB seeds were sorted to remove extraneous materials. The seeds were soaked for 12h, it was rinsed and heated for 5min at 100ºC and left to cool. The seeds were manually dehulled and blended with water in the ratio 1:8 w/v until a smooth slurry was obtained. The slurry was filtered through a double folded muslin cloth and then pasteurized at 65ºC for 30min, it was cooled to 25ºC, bottled in a glass container and then stored in the refrigerator for further usage (Taiwo and Zulfah, 2014).

Inoculum preparation for fermentation

The inoculum (identified by Bioinformatics services, Ibadan, Nigeria) was prepared using the method described by Obinna - Echem (2018). A distinct colony from the agar plate culture was inoculated into 10ml of broth incubated at 37ºC for 18-20h. The culture was harvested by centrifugation at 4000rpm for 10min and washed twice in phosphate buffered saline (PBS) (pH7.3±0.2). Such that 1ml of inoculum will produce 9 and 8 $\log_{10}$ cfu/ml. 5ml of the washed isolate which was suspended in sterile distilled water was inoculated into 100ml of the AYB and malted red rice milk and allowed to incubate at 37ºC for 24h which was used as the stock starter culture.

Optimization of AYB/ MRR yoghurt analogues

The optimization of the mixture and process (inoculum) components was performed using combined optimal process (I-Optimal) design in response surface methodology (RSM) to find the optimum constituents for the fermentation, no blocks with twenty eight runs was generated, as shown in the design matrix (Table 1). the mixture components (A, B and C) were coded low and high, with values ranging from 0.71-0.99, 0-0.289 and 0.001-0.01 for African yambean, red rice and xanthan gum respectively, as shown in the mixture component (Table 1) variables viscosity, syneresis, and acceptability was analysed as responses (Table 1). The process factor was represented as D. All experiments were performed in triplicates. Optimization was performed using design expert (Stat-Ease Inc., Minneapolis, MN, USA) software version11 (Stat-Ease, 2018).

Table 1: Combined optimal design matix of AYB/MRR yoghurt analogues and their responses

Run	Component 1 A:AYB	Component 2 B:MRR	Component 3 C:XAN	Factor 4 D:INOCULUM Ml	Viscosity (pa.s)	Syneresis (%)	Acceptability
1	0.728476	0.261524	0.01	14.8			
2	0.988045	0.00886294	0.00309193	14.6			
3	0.711853	0.282427	0.00572012	9.9			
4	0.74153	0.25747	0.001	5			
5	0.865809	0.127941	0.00624984	15			
6	0.71	0.289	0.001	12.4			
7	0.82037	0.173697	0.00593257	5			
8	0.725936	0.269856	0.00420804	15			
9	0.890475	0.104718	0.00480653	10			
10	0.893324	0.0966759	0.01	7.4			
11	0.865809	0.127941	0.00624984	15			
12	0.806891	0.183109	0.01	12.5			
13	0.99	0	0.01	9.65			
14	0.71	0.28	0.01	5			
15	0.908697	0.0813026	0.01	12.5			
16	0.831774	0.158226	0.01	5			
17	0.95188	0.0410797	0.00704046	5			
18	0.83932	0.15968	0.001	15			
19	0.890475	0.104718	0.00480653	10			
20	0.711853	0.282427	0.00572012	9.9			
21	0.770118	0.228882	0.001	10.05			
22	0.777363	0.212637	0.01	7.4			
23	0.954682	0.0443178	0.001	9.5			
24	0.866071	0.132929	0.001	5			
25	0.929952	0.0600475	0.01	15			
26	0.890475	0.104718	0.00480653	10			
27	0.82037	0.173697	0.00593257	5			
28	0.807003	0.182997	0.01	9.95			

Physicochemical evaluation of optimized yoghurt analogues from African

Determination of viscosity for African yambean/ malted red rice yoghurt analogues

The method of Barber *et al.* (2021) was used to determine the viscosity of the samples. Each of the yoghurt samples (200 ml) was homogenized separately in a homogenizer (FJ 300-S China) at medium speed for 3min. The viscosity of the thoroughly homogenized samples were measured using a digital display viscometer (NDJ-85, China) with No. 4 spindle at 120 rpm. Fifty (50) ml of each sample was introduced into clean dried viscosity tube. The viscometer was placed into a holder and the sample temperature was set to the bath temperature of 30ºC for 30min. The afflux time was recorded by timing the flow of the same as it flows freely from the upper timing mark back to the lower timing mark. The viscosity was determined in centipoise (cp) and calculated as:

Viscosity (V) = C× t;

Where V = Viscosity at 30ºC, t = time (s), C = viscosity tube constant (0.09757)

Determination of syneresis for African yambean/ malted red rice yoghurt analogue

The method of Barber *et al* (2021) was used to measure this parameter. Twenty milliliter (20 ml) of each of the yoghurt formulations (20 ml) was centrifuged (L-600 China centrifuge) at 5000rpm for 10 min. Syneresis Index (SI) in percentage was calculated as:

$$\% \text{ Syneresis} = \frac{Volume\ of\ supernatant}{Weight\ of\ sample} \times 100$$

Acceptability of AYB/MRR yoghurt analogues (optimized runs)

The degree of likeness for the yoghurt analogue was determined using the method described by Iwe (2010). The samples were presented to semi-trained ten-member panel who were familiar with yoghurt. The panelists were provided with 20ml of each yoghurt analogue sample. The samples were coded and presented to the panelists using white glass cups. Water was provided for mouth wash in between evaluations. Panelists rated the yoghurt based on the sensory attributes of colour, appearance, consistency, aroma, taste and mouthfeel using the 9-point hedonic scale. 1: dislike extremely, 2: dislike very much, 3: dislike moderately, 4: dislike slightly, 5: neither like nor dislike, 6: like slightly, 7: like moderately, 8 like very much and 9: like extremely.

Results and Discussion

ANOVA and fit statistics for viscosity of AYB/MRR yoghurt analogues, based on response surface methodology

The analysis of variance and fit statistics for viscosity of fermented AYB, MRR and Xanthan gum blends is presented in Table 2. The model was significant ($p<0.05$) with p-value <0.0001. This finding is in agreement with the report of Nehaa *et al.* (2022) who reported a significant effects of coconut, oats and peanut milk on the yoghurt viscosity. Coefficient of determination (R^2), Adjusted R^2 and Predicted R^2 were respectively 0.9992, 0.9969 and -0.6228, while the coefficient of variability (C.V) and adequate precision were respectively 0.992 % and 129.0787. Mean viscosity was 2.09. Lack of fit, with p-value of 0.3246 was not significant ($p>0.05$). The goodness-of-fit of the model was ascertained by the coefficient of determination (R^2). It is a measure of the amount of variation around the mean explained by the model (Stat-Ease, 2018). The best R^2 value for a good model fitting was estimated between 0.8 and 1.0 (Jusoh *et al*., 2013). Consequently, R^2 of 0.9992 (99.92 %) given for viscosity indicates good fit for the model. The R^2 predicted will decrease when there are too many insignificant values in the model (Ghosh *et al*., 2012). As per thumb rule, these values should be within 0.2 of each other. Negative predicted R^2 (-0.6228) indicates more terms that were insignificant (Ghosh *et al*., 2012).

Table 2: ANOVA and fit statistics for viscosity of AYB/MRR yoghurt analogues.

Source	Sum of Squares	Df	Mean Square	F-value	p-value	
Model	3.69	20	0.1846	429.93	< 0.0001	significant
(1)Linear Mixture	0.4175	2	0.2087	486.05	< 0.0001	
AB	0.6408	1	0.6408	1492.12	< 0.0001	
AC	0.0506	1	0.0506	117.81	< 0.0001	
AD	0.0105	1	0.0105	24.51	0.0017	
BC	0.0670	1	0.0670	156.01	< 0.0001	
BD	0.0319	1	0.0319	74.19	< 0.0001	
CD	0.0084	1	0.0084	19.53	0.0031	
ABC	0.3171	1	0.3171	738.34	< 0.0001	
ABD	0.0103	1	0.0103	23.91	0.0018	
ACD	0.0054	1	0.0054	12.62	0.0093	
BCD	0.0078	1	0.0078	18.06	0.0038	
AD^2	0.1391	1	0.1391	323.89	< 0.0001	
BD^2	0.0024	1	0.0024	5.64	0.0493	
CD^2	0.0567	1	0.0567	131.95	< 0.0001	
ABCD	0.0131	1	0.0131	30.43	0.0009	
ABD^2	0.2554	1	0.2554	594.62	< 0.0001	
ACD^2	0.0436	1	0.0436	101.51	< 0.0001	
BCD^2	0.0539	1	0.0539	125.40	< 0.0001	
$ABCD^2$	0.1285	1	0.1285	299.26	< 0.0001	
Residual	0.0030	7	0.0004			
Lack of Fit	0.0011	2	0.0005	1.42	**0.3246**	not significant
Pure Error	0.0019	5	0.0004			
Cor Total	3.70	27				
Fit Stat.						
R^2	0.9992					
Adj. R^2	0.9969					
Pred. R^2	-0.6228					
CV	0.992%					
Adeq. Prec.	129.0787					
Mean	2.09					
Std Dev.	0.0207					

$P<0.05$ (significant), **$p>0.05$** (not significant)

The estimated regression coefficient and interaction effect of AYB, MMR, xanthan gum and inoculum size on viscosity during yoghurt fermentation is presented in equation 1 and the 3D surface plot of Figures 1 and 2. The estimated regression coefficient for viscosity showed that all linear mixture terms were significant ($p<0.05$). Result also showed that A, B, AC, AD, BC, BD, ABC, ACD, CD and CD^2 are significant model terms and synergistic to viscosity of the fermented mixture. C, AB, CD, ABD, AD^2 and BD^2 are also significant model terms, but antagonistic to the viscosity. Increased interaction effect of

ABD showed a synergistic effect on the viscosity (Fig. 2). The fitted regression model in terms of coded factors, excluding insignificant terms:

$$\textbf{Viscosity} = +5.52A + 2.05B - 802.59C - 10.38AB + 726.33AC + 1.13AD + 820.84BC - 2.87ABD + 278.33ACD + 361.92BCD - 4.42AD^2 - 0.1742BD^2 + 1212.89CD^2 \quad \ldots..(1).$$

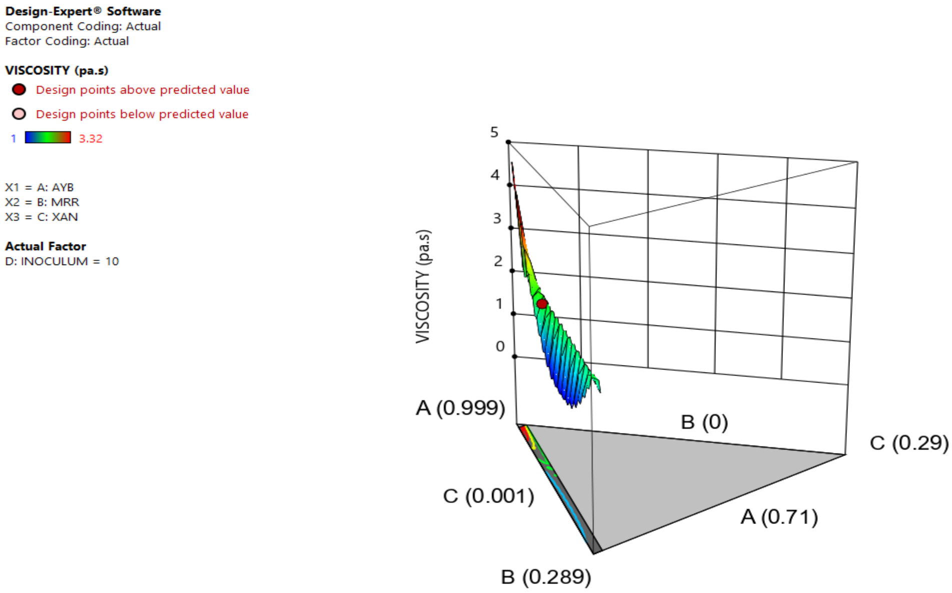

Figure 1: Effects of interaction of AYB, MRR and XAN on Viscosity of AYB/MRR yoghurt analogues

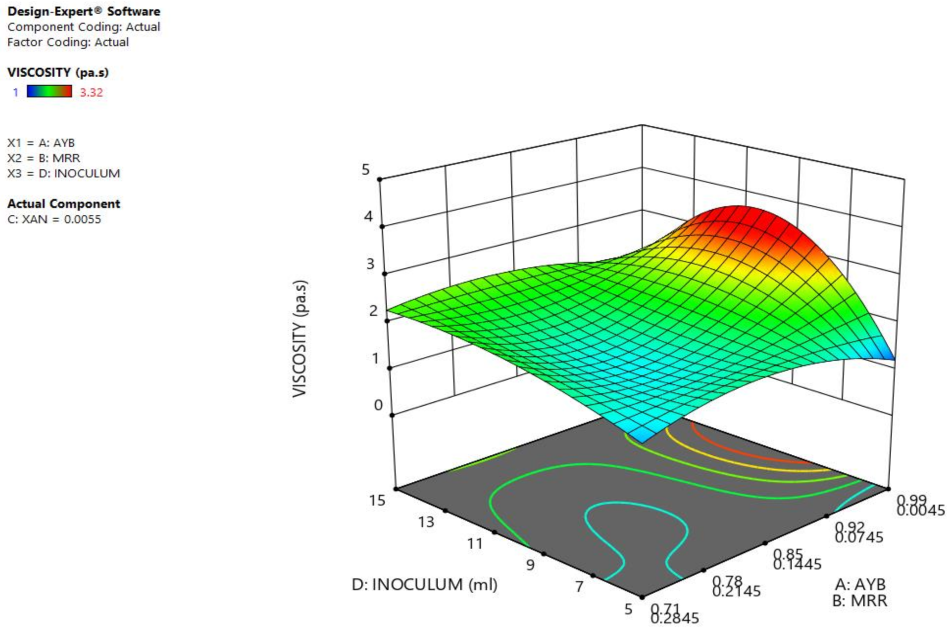

Figure 2: Effects of interaction of AYB, MRR and inoculum on viscosity of AYB/MRR yoghurt analogues.

ANOVA and fit statistics for syneresis of AYB/MRR yoghurt analogues

The Analysis of variance, and fit statistics for syneresis of fermented AYB, MRR and Xanthan gum blends (Table 3) showed that the model was significant ($p<0.05$) with p-value of <0.0001. Coefficient of determination (R^2), Adjusted R^2 and Predicted R^2 were respectively 0.9983, 0.9975 and 0.9954, while the coefficient of variability (C.V) and adequate precision were respectively 6.58 % and 89.6151. Lack of fit, with p-value of 0.1708 was not significant ($p>0.05$).
.

Table 3: ANOVA and fit statistics for syneresis of AYB/MRR yoghurt analogues

Source	Sum of Squares	Df	Mean Square	F-value	p-value	
Model	11243.11	9	1249.23	1183.53	< 0.0001	significant
[(1)]Linear Mixture	7666.71	2	3833.36	3631.75	< 0.0001	
AB	109.36	1	109.36	103.61	< 0.0001	
AC	40.08	1	40.08	37.98	< 0.0001	
BC	40.63	1	40.63	38.49	< 0.0001	
ABC	42.17	1	42.17	39.95	< 0.0001	
AB(A-B)	6.90	1	6.90	6.54	0.0198	
AC(A-C)	41.23	1	41.23	39.06	< 0.0001	
BC(B-C)	43.00	1	43.00	40.74	< 0.0001	
Residual	19.00	18	1.06			
Lack of Fit	16.38	13	1.26	2.40	**0.1708**	not significant
Pure Error	2.62	5	0.5245			
Cor Total	11262.11	27				
Fit Stat.						
R^2	0.9983					
Adj. R^2	0.9975					
Pred. R^2	0.9954					
CV	6.58%					
Adeq. Prec.	89.6151					
Mean	15.62					
Std Dev.	1.03					

P<0.05 (significant), **p>0.05** (not significant).

The estimated regression coefficient and interaction effect of AYB, MMR, xanthan gum and inoculum size on Syneresis during yoghurt fermentation is presented in equation 3.2 and the 3D surface plot of Figure 3. The estimated regression coefficient for syneresis showed that all linear mixture terms were significant ($p<0.05$). Result also showed that A, B, AB, AC, BC, are significant model terms and synergistic to syneresis of the fermented mixture. C, ABC, are also significant model terms, but antagonistic to the syneresis. Increased interaction effect of ABC showed an antagonistic effect on the syneresis.

The fitted regression model in terms of coded factors, excluding insignificant terms:

Syneresis = +28.99A + 56.79B – 0.000026C + 45.34AB + 0.00004AC + 0.0000408BC – 0.000028ABC + 12.68AB(A-B) – 0.000044AC(A-C) – 0.000044BC(B-C) …..(2).

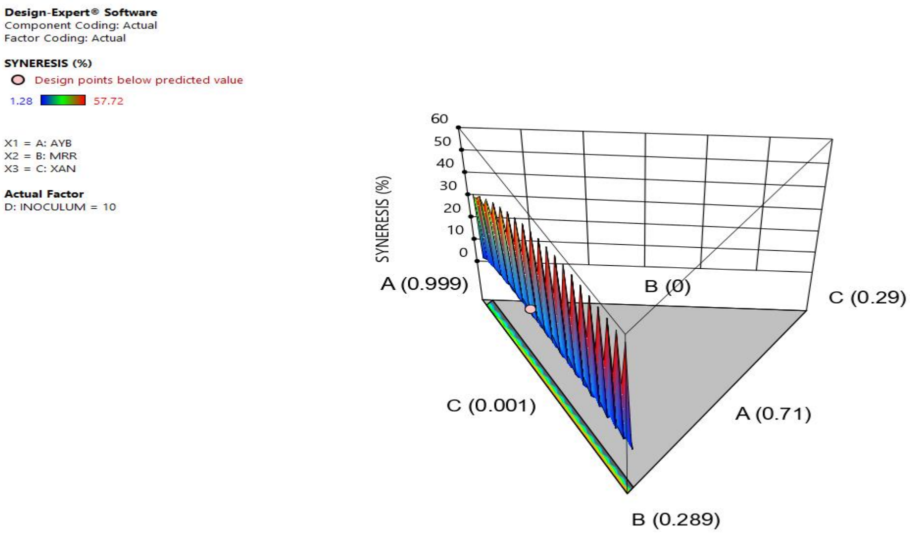

Figure 3: Effects of interaction of AYB, MRR and XAN on syneresis of AYB/MRR yoghurt analogues.

ANOVA and fit statistics for acceptability of AYB/MRR yoghurt analogues blends

The analysis of variance, and fit statistics for Acceptability of fermented AYB, MRR and Xanthan gum blends (Table 4) showed that the model was significant ($p<0.05$) with p-value of <0.0001. Coefficient of determination (R^2), Adjusted R^2 and Predicted R^2 were respectively 0.9904, 0.9677 and 0.9677, while the coefficient of variability (C.V) and adequate precision were respectively 6.39 % and 23.88. Lack of fit was not significant ($p>0.05$). CV is a measure of deviation from the mean values, which shows the reliability of the experiment (Iwe *et al*., 2023). It is the standard deviation expressed as a percentage of the mean. CV also describes the extent to which the data were dispersed as well as the reproducibility and repeatability of the model (Firatiligil-Durmus and Evranus, 2010). Shishir *et al*. (2016) reported that a CV < 10% indicates better precision and reliability. The goodness-of-fit of the model was also ascertained by the coefficient of determination (R^2). It is a measure of the amount of variation around the mean explained by the model (Stat-Ease, 2018). Jusoh *et al.* (2013) reported that the best R^2 value for a good model fitting was estimated between 0.8 and 1.0. Consequently, R^2 of 0.9904 (99.04%) given for acceptability indicates good fit for the model. Adequate precision was 22.88. Adequate precision measures the signal to noise ratio. It compares the range of the predicted values at the design points to the average prediction error. A ratio greater than 4 is desirable (Edem and Elijah, 2016; Stat-Ease, 2018). The ratio of 22.88 given, indicates an adequate signal. The model can be used to navigate the design space.

Table 4: ANOVA and fit statistics for acceptability of AYB/MRR yoghurt analogues.

Source	Sum of Squares	Df	Mean Square	F-value	p-value	
Model	63.45	19	3.34	43.56	< 0.0001	Significant
(1)Linear Mixture	18.04	2	9.02	117.65	< 0.0001	
AB	1.03	1	1.03	13.43	0.0064	
AC	0.0049	1	0.0049	0.0643	**0.8062**	
AD	1.44	1	1.44	18.75	0.0025	
BC	0.0053	1	0.0053	0.0690	**0.7994**	
BD	0.9779	1	0.9779	12.76	0.0073	
CD	1.55	1	1.55	20.22	0.0020	
ABC	0.0054	1	0.0054	0.0699	**0.7981**	
ABD	1.45	1	1.45	18.88	0.0025	
ACD	1.55	1	1.55	20.23	0.0020	
BCD	1.55	1	1.55	20.23	0.0020	
AB(A-B)	0.4610	1	0.4610	6.01	0.0398	
AC(A-C)	0.0048	1	0.0048	0.0627	**0.8086**	
BC(B-C)	0.0060	1	0.0060	0.0781	**0.7869**	
ABCD	1.55	1	1.55	20.24	0.0020	
ABD(A-B)	0.6358	1	0.6358	8.29	0.0205	
ACD(A-C)	1.55	1	1.55	20.24	0.0020	
BCD(B-C)	1.55	1	1.55	20.23	0.0020	
Residual	0.6133	8	0.0767			
Lack of Fit	0.2671	3	0.0890	1.29	**0.3749**	not significant
Pure Error	0.3462	5	0.0692			
Cor Total	64.06	27				
Fit Statistics						
R2	0.9904					
Adj. R2	0.9677					
Pred. R2	0.9677					
C.V	6.39					
Adeq. Prec.	23.88					
Mean	4.33					
Std. Dev.	0.277					

P<0.05 (significant), **p>0.05** (not significant)

The estimated regression coefficient and interaction effect of AYB, MMR, xanthan gum and inoculum size on acceptability during yoghurt fermentation is presented in equation 3 and the 3D surface plot of Figure 4. The estimated regression coefficient for viscosity showed that all linear mixture terms were significant ($p<0.05$). Result also showed that A, B, AC, AD, BC, BD, ABC, ACD, CD and CD^2 are significant model terms and synergistic to viscosity of the fermented mixture. C, AB, CD, ABD, AD^2 and BD^2 are also significant model terms, but antagonistic to the viscosity. Increased interaction effect of ABD showed a synergistic effect on the viscosity (Fig. 4).

The fitted regression model in terms of coded factors, excluding insignificant terms:

Acceptability = + 7.53A – 1.29B + 0.000014C + 9.53AB + 79.48AD + 4.73BD – 168.13ABD …..(3).

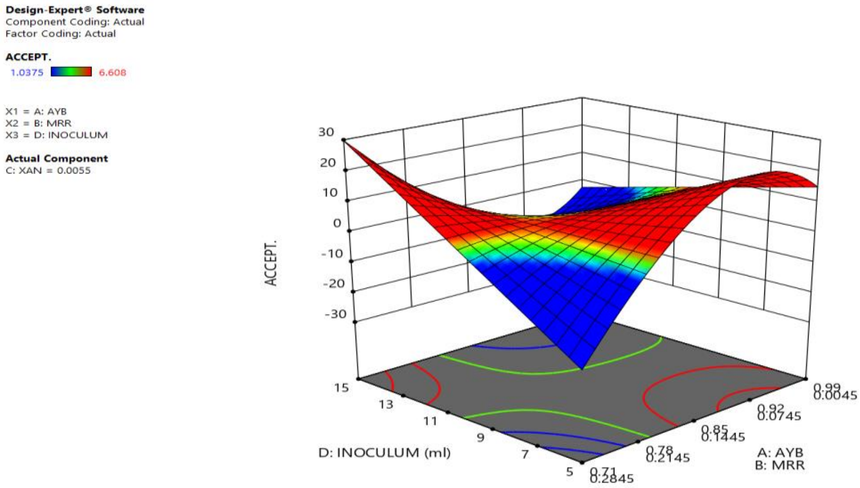

Figure 4: Effects of interaction of AYB, MRR, XAN and inoculum on acceptability of AYB/MRR yoghurt analogues.

Conclusion

Yoghurt analogues were produced using African yambean (*Sphenostylis stenocarpa Hams*) and malted red rice (*Oryza glaberrima*). Optimization of the mixture components and process factor was carried out using combined optimal mixture design, viscosity, syneresis and acceptability as responses were analysed. The suitable mathematical models developed for the optimization of the fermentation variables in African yambean and malted red rice yoghurt is highly recommended for the development of a novel non-dairy probiotic yoghurt.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests nor personal relationships that could have appeared to influence the work reported in this paper.

References

Abioye, V. Olanipekun, B., & Omotosho, O., (2015). Effect of varieties on the proximate, nutritional and anti-nutritional composition of nine variants of African yam bean seeds (*Sphenostylis stenocarpa*). *Donnish J. Food Sci. Technol.* 1,17-21.

Adeomowaye, B.I.O. Tucker, G.A. & Smetanska, I., (2015). Nutritional potential of nine underexploited legumes in Southwest Nigeria. *Int, Food Res. J.* 22, 798-806.

Ajibola, G.O. & Olapade, A.A., (2016). Physical, proximate and anti-nutritional composition of African yam bean (*Sphenosstylis stenocarpa*) seeds varieties. *J. Food Res*. 5, 67.

Anya, M.I. & Ozung, P.O., (2019). Proximate, mineral and anti-nutritional compositions of raw and processed African Yam bean (*Sphenosstylis stenocarpa*) seeds in Cross River State, *Nigeria Global J. Agric. Sci*. 18, 19.

AOAC, (2010) Official Methods of Analysis of Association of Official Analytical Chemists. 18th Edition, Washington, DC.

Baiyeri, S. Uguru, M. Ogbonna, P., Samuel-Baiyeri, C.C. Okechukwu, R, Kumaga, F., & Amoatey, C., (2018). Evaluation if the nutritional composition of the seeds of some selected African yam bean (*Sphenosstylis stenocarpa*). *Agro-Sci. J. Trop.Agric. Food Environ. Ext*. 17, 37-44.

Barber, L.I., Onuegbu, N.C., Osuji, C.M. & Ogueke, C.C. (2021). Storage stability of probiotic soy yoghurts with enzyme hydrolysed African breadfruit and rice additives. *European Journal of Nutrition & Food Safety, 13*(2), 110-121.

Cokro Wlnijaya & Andreas Romulo (2012). Proximate analysis and antioxidant activity of red rice (oryza sativa L.) milk. *Journal of Physics: Conference Series* 10:1088 – 1742.

Daniela, C.S., Leandro, T.B., Luiz, A.M. & Beatrize, O.S. (2016). Post-acidification and evaluation of anthocyanins stability and antioxidant activity in fermented milk and yoghurts. *Rev. Bras. Frutic 39* (5) 871.

Dennis, A., Savariano, R. & Hutkins, W. (2020). Yoghurt, cluttered fermented milk, and health: A systematic review. Nutrition review vo. 79:599-614.

Duodu, K.G., & Apea-Bab, F.B., (2017). African Legumes Nutritional and Health Promoting Attributes Gluten-free Ancient Grains: Cereals, Pseudocerals, and Legumes: Sustainable, Nutritional, and Health Promoting Foods for the 21st Century, Elsevier Ltd.

Edem, V.E. and Elijah, A. (2016). Optimization of Coconut (Cocos nucifera) Milk Extraction Using Response Surface Methodology. International Journal of Nutrition and Food Sciences, 5 (6): 384-394.

Edith Ani, Julius Amore, Bibioma Igbabu (2018). Physiochemical, microbiological, sensory properties and storage stability of plant-based yoghurt produced from Banbaramal, soybean and moringa deifera seed milks. *American Journal of Food and Nutrition, 6*(4), 115-125.

El-Kholy, A.M., Osman, M., Gouda, A. & Ghareeb, W.A. (2011). Fortification of Yoghurt with Iron.*World Journal of Dairy & food Sciences*, **6(2)**: 159-165.

Erdogan Kocukoner & Zekai Ta Rakci (2005). Influence of different fruit additives on some properties of stirred yoghurt during storage. *World Journal of Dairy & food Sciences*, **4(1)**: 59-65.

Firatiligil-Durmus, E., and Evranuz, O. (2010). Response surface methodology for protein extraction, optimization of red pepper seed (Capsicum frutescens). *LWT-Food Science and Technology, 43* (2): 226-231.

Ghosh, S., Chakraborty, R. and Raychaudhuri, U. (2012). Optimizing process conditions for palm (Borassus flabelliffer) wine fermentation using Response Surface Methodology. *International Food Research Journal 19*(4): 1633-1639.

Iwe, M.O. (2010). Handbook of Sensory Methods and Analysis. 2nd Edition, Rojoint Communication services Ltd., Enugu. Pp 75-78 *Journal of Food Science*, 12(4): 73-79.

Iwe, M.O., Chibor, B.S. and Obasi, N.E. (2023). Optimization of the Chemical Interesterification of Allanblackia (Allanblackia floribunda), African Walnut (Tetracarpidium conophorum) and Fluted Pumpkin (Telfairia occidentalis) Seed Oil Blend for use in Bakery Shortening Production, a Response Surface Approach. Retrieved from www.researchgate.net October 2023.

Jusoh, J. M., Rashid, N. A. and Omar, Z. (2013). Effect of sterilization process on deterioration of bleachability index (DOBI) of crude palm oil (CPO) extracted from different degree of oil palm ripeness. *International Journal of Bioscience, Biochemistry and Bioinformatics, 3* (4): 322-327.

Klu, G.Y., Bansa, D., Kumaga, F.K. Aboagye, I.M. Benett-Lartey, S.O., & Gamedogbao, D.K. (2000). The African yam bean (*Sphenosstylis stenocarpa*): neglected crop in Ghana West Afr. *J. Appl. Ecol*.

Kolawole, O., Falode, Opeolu M., Ogundele Adenike, O., Ogunlsue Olanrewaju, E., Fayemi Fidelis, C.K. (2015). Physio-chemical, sensory and microbiological characteristics of palm yoghurt from bambaran groundnut and soybean. *Journal of Food Science Technology, 52*(9), 5858-5865.

Kumar, H., Salminen, S., Verhagen, H., Rowland, I., Banares, S., Young T., Nomoto K., Lalonde (2015). Novel probiotics and prebiotics: Road to the market. *Curr. Opin Biotechnol* 32, 99-103.

Kunze, W (2005). Technology of brewing and malting. VLB, Berlin. Pp.110.

Mbaeyi, I.E. (2011). Studeis on cereal legume blend potential for development of novel non-dairy probiotic yoghurt analogues. Ph.D Thesis submitted to the Department of Microbioogy Faculty of Biological Sciences, University of Nsukka, Nigeria. 1-222.

Nehaa Baskar, Sabitha Varadharajan, Mathushree Rameshbabu, Sudha Ayyasamy and Sangeetha Velusamy (2022). Development of plant – based yoghurt. *J. Foods and Raw Materials, 10*(2).

Ndidi, U.S. Ndidi, C.U. Olagunju, A. Muhammad, A. Billy, F.G. & Okpe, O. (2014). Proximate, ant nutrients and mineral composition of raw and processed (boiled and roasted) *Sphenosstylis stenocarpa* seeds from Southern Kaduna, Northwest Nigeria ISRN *Nutr*.1-9.

Nwosu, J.N. (2013). Evaluation of the proximate composition and anti-nutritional properties of African yam bean (*Sphenosstylis stenocarpa*) using malting treatment.*Int. J. Basic Appl. Sci*. 2, 157/169.

Obinna-Echem, P. C. (2018). Acid, Bile and Aggregation Abilities of *Lactobacillus plantarum* Strains Isolated from Akamu a Nigerian Fermented Maize Food. *American Journal of Food Science and Technology, 6* (1), 7-11.

Okeke, P.N. & Chikwendu, J.N. (2015). Effects of varying fermentation periods on the chemical composition of African yambean (*Sphemostylis stenocarpa*) and Aclua (*Digitaria exilis*) flour blends and sensory properties of their products, International Scholarly and Scientific Research & Innovation. 9(4), 431 – 438.

Rathna Priya T.S., Ann Raeboline Lincy Eliazer Nelson, Kavitha Ravichandran & Usha Anthony (2019). Nutritional and functional properties of coloured rice varieties of south India: a review *Journal of Ethnic Foods* 6:11. Rood, M.A. (2000). Red menace. *Rice Journal*, 103 (March): 18-20.

Rood, M.A. (2000). Red menace. Rice Journal 103 (March): 18-20.

Sangkitikomon, V., Tentumnon, T. & Rodelnanasasod, A. (2008). Comparisions of total antioxidants of red rice, black rice and black sticky rice. *Journal of Nutrient*, 213, 16-21.

Shishir, M. R. I., Taip, F. S., Aziz, N. AB., Talib, R. A. and Sarker, Md. S. H. (2016). Optimization of spray drying parameters for pink guava powder using response surface methodology. Food Science and Biotechnology Journal, 25 (2): 461- 468.

Soomro, A.H., Masud, T. & Kiran Anwaar (2002). Role of lactic acid bacteria (LAB in food preservation and human health. A review, *Pakistan Journal of Nutrition* 1(1): 20-24.

Stat-Ease (2018). Design-Expert.www.stateease.com

Taiwo, A. Aderinola & Zulfaln Y. Olanrewaju (2014). Quality and microbiological evaluation of African yam bean yoghurt supplemented with coco milk. *Journal of Microbiology, Biotechnology and Food Science, 4*(1), 70-74.

Toyosi, T., George, Anthony, O. Obilana, Samson, A. & Oyeyinka (2020). The prospects of African yam bean: Post and future importance. Heliyon 6: 05458.

Uzuner, E.A., Ozer, K., Figen, K., Gamze, Y. & Oktay, Y. (2016). Usage of rice milk in probiotic yoghurt production. *Carpathian J. of food Sci and Tech*. *8*(4) 5-24.

Zanhi, N.K. & Jidemi, I.A. (2012). Physico-chemical and sensory qualities of soy and milk solids fortified low fat yoghurt. *African Journal of Agric. Research, 7*(38), 5336-5343.

•*Thank you for publishing with us.*