



Kinetic acidification and quality composition of yoghurt produced with soursop puree

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Abstract

The growing demand for healthy yoghurt products necessitates the need to investigate the effect of process parameters and soursop puree to sugar concentration on the kinetic acidification, quality composition and sensory attributes of yoghurt. Taguchi design was used to interact incubation temperature (39–43 °C), pasteurization temperature (80–100 °C) and soursop puree to sugar ratio (0%, 50% and 75%) to produce nine yoghurt samples. The acidification kinetics of the yoghurt samples were estimated using maximum acidification rate, time required to achieve maximum acidification and time at which pH of 4.6 was reached. The proximate composition, mineral content (potassium, calcium and phosphorus), vitamin C, total phenolic content were the quality composition that were determined while the sensory attributes of the yoghurt samples were also evaluated. The maximum acidification rate, time required to achieve maximum acidification and time at which pH of 4.6 was reached were 0.0042 pH/min, 2 h and 3.5 h, respectively. This occurred when 75% of soursop puree to sugar ratio was used to produce yoghurt. The moisture content of the yoghurt samples decreases as the soursop to sugar ratio increases. The crude protein, crude fat, carbohydrate, fibre, vitamin C and total phenolic contents increases as the soursop to sugar ratio increases. The calcium content of the yoghurt samples ranges from 298.5 to 623.5 mg/l, 408.5 to 810.5 mg/l in potassium and 120–519 mg/l in phosphorous, respectively. This information could assist yoghurt producers to develop new yoghurt varieties with improved quality and sensory profile.

Keywords Kinetic acidification · Yoghurt · Soursop · Quality composition

Introduction

Yoghurt is a fermented milk product obtained by the lactic acid fermentation through the incorporation of *Streptococcus salivarius* subsp. *Thermophilus* and *Lactobacillus delbrueckii* subsp. *Bulgaricus*. The milk coagulates when enough quantity of lactic acid is produced, and this coagulated milk is called yoghurt. Yoghurt is a dairy food product that is widely known and appreciated by its consumer throughout the world. It is widely available in various forms such as; textures (i.e., liquid, set, smooth), fat contents (luxury, low-fat, virtually fat-free) and flavours (natural, fruit, cereal) [1]. The decrease in the pH of milk

during fermentation is called “acidification of milk” and this process is one of the oldest methods of preserving milk [2]. According to Medeiros et al. [3], the biochemical process which brings the particular yoghurt identity where the starter culture produces lactic acid, aromatic and volatile compounds is known as acidification process. Yoghurt can be categorized into two groups and these are standard culture yoghurt and bio yoghurt or probiotic yoghurt [4]. Standard yoghurt is manufactured from the conventional starter culture strains, *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus acidophilus* subsp. *thermophilus* [5]. Meanwhile, bio-yoghurt or probiotic yoghurt is supplemented with probiotic strains such as *Bifidobacterium* and *Lactobacillus acidophilus* that are proven to have many health benefits and should remain live at adequate numbers [1].

Yoghurt supplies good quality proteins and is also an excellent source of calcium, phosphorus, potassium and contains significant quantities of vitamins B₆ and B₁₂ [2]. Yoghurt has many health benefits other than its rich nutritional profile; example is the case of lactose intolerance

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where it attains all the benefits of milk products without causing discomforts associated with hypolactasia [2]. The consumption of fermented dairy products containing probiotic bacteria can also provide numerous advantages. A high population of probiotic organisms in the colon can contribute to good intestinal health. Consequently, consuming yoghurt that contains viable probiotic organisms adds benefit to human gut health. According to Afzaal et al. [5], the inclusion of the probiotic bacteria in the yoghurt samples either in encapsulated state or without encapsulation significantly affected syneresis, lactose, acidity, pH and viscosity. Functional foods are food products mainly designed to have advance benefits that are more than the original nutritional contents [6]. Functional food products are becoming part of consumers' everyday lives because of the healthy benefits and as well as increased scientific evidence of their effectiveness among consumers [7]. In recent years, studies have focused on improving yoghurt quality, including sensory quality and functional properties. The addition of synthetic compounds to yoghurt has been reported to have negative effect to the nutritional value of yoghurt due to their potential toxicity to human body. Thus, many efforts have been made to find safe supplements from natural sources [8]. Consumers tend to choose flavour as a key factor in food for acceptance apart from their health benefits, that is why there has been attempts to improve yoghurt functionality by addition of different fruits [1]. Salami et al. [9] incorporate Zucchini to Yoghurt and it was observed that the addition increases the health functionalities of the yoghurt. Pérez-Chabela et al. [10] produce functional yoghurt by fortification with mango peel and potato peel flour. Therefore, due to the present health crisis in the world, health conscious consumers of yoghurt tend to prefer yoghurt that would contain one or more health-promising properties beyond the traditional yoghurt nutrients.

Soursop (*Annona muricata* L.) is one of the underutilized fruit in Nigeria but it is known to have a good nutritional profile. Soursops (*Annona muricata* L.) are fruits which when ripe are highly perishable as become soft and easily bruised [11]. Soursop gives a custard flavour when ripe which has a pleasant aroma [12]. The puree when consumed is sweet, juicy and creamy. Its unique flavour contributes to the acceptance of its products in the international market [12]. The soursop puree is an excellent source of vitamins B and C, potassium, riboflavin and niacin [13]. Soursop fruit contains 62 to 85% puree, 25 to 30% skin and 2 to 3% seed on weight basis [13]. The fruit puree is thick and requires dilution before use as juice or squash. The fruit can also be used to make an excellent jam or jelly. The soursop has some medicinal uses such as remedy for gall bladder infection, coughs, diarrhoea, dysentery and indigestion. Nwae-hujor [14] reported that soursop has anti-cancerous properties. The white edible puree of soursop contains 80–81%

water, 1% protein, 18% carbohydrate, 3.43% titratable acidity, 24.5% non-reducing sugar, and vitamins B₁, B₂, and C [15]. Therefore, there is need to explore the usefulness of soursop by incorporating it into yoghurt with the aim of producing improved quality yoghurt. Lutchmedial et al. [11] incorporated soursop nectar at 0%, 5%, 10% and 15% to produce stirred yoghurt with improved protein and mineral content. Senadeera et al. [16] evaluated the addition of soursop puree of *A. muricata*, *A. squamosa* and *A. reticulate* on physicochemical and microbiological properties of probiotic yoghurts. Chang et al. [17], produced dried soursop powder from soursop puree while Pham et al. [18] evaluated the effect of fermentation by the addition of soursop puree into condensed milk and cow milk. Loveday et al. [19] reported that milk composition, thermal treatment and incubation temperature could influence the acidification process and characteristics of the yoghurt formation. However, the intensity at which the factors would affect the acidification process and the quality composition of yoghurt produced from soursop is sparse in the literature. Therefore, the objective of this study was to evaluate the effect of pasteurization temperature, incubation temperature and soursop puree to sugar ratio on the kinetic acidification, quality composition and sensory attributes of yoghurt samples using Taguchi design.

Materials and methods

Materials

The Soursop fruits (*Annona muricata* L) used for this study were purchased from Harmony Farms (Ilorin, Nigeria), skimmed milk powder was purchased from Dano Milk, Arla Foods (Lagos, Nigeria), sugar was purchased from Dangote sugar (Ilorin, Nigeria) and freeze-dried yogurt starter culture powder for yoghurt fermentation (Nigeria) contains *Lactobacillus bulgaricus*, *S. thermophilus*, and *L. acidophilus*.

Preparation of soursop puree

Fresh soursop fruit (*Annona muricata* L) was washed, weighed, peeled with sharp knife and de-seeded. The remaining mesocarp of the fruit was weighed to determine the amount of water to be added. The average weight of the mesocarp obtained was the amount of water added to the mesocarp. The mesocarp and the water were blended using a blender (Sonik SB-736, Japan) to obtain a thick and creamy colour soursop puree.

Experimental design

Nine (9) samples of yoghurt were prepared using Taguchi experimental design at three independent variables and

levels as shown in Table 1. The independent variables of the design were pasteurization temperature (80 °C, 90 °C and 100 °C), inoculation temperature (39, 41 and 43 °C) and concentration of soursop puree to sugar ratio (0, 50% and 75%), respectively. The yoghurt samples with 0% soursop puree are plain yoghurt.

Preparation of yoghurt samples

Potable water of 30% by mass was introduced in a pasteurizing vat and was heated to temperature of 80, 90 and 100 °C, respectively. The temperature was monitored and measured with the aid of an infrared thermometer (IRT 206, Taiwan). As soon as the desirable temperature was reached, 24% by mass of the concentration of sugar to soursop puree was divided into 0%, 50%, and 75% and were added to the heated water. i.e. 0% means no soursop puree or plain yoghurt,

50% means, 50% of sugar and 50% of soursop puree while 75% means, 75% of soursop puree and 25% of sugar. The skimmed milk (45% by mass) was also introduced to the mixture. The mixture of dissolved skimmed milk, sugar and soursop puree were thoroughly stirred together at the pasteurization temperatures for 20 min using a hand mixer (TB-HM009, CUSINAID) to get homogenized mixture. The mixture was allowed to cool to 45 °C inside the pasteurizing vat and thereafter, 1% of starter culture was added to the mixture and stirred thoroughly to enable even distribution of the culture. The mixture was filled into sterilized polypropylene bottles and placed inside an apparatus for conditioning dough and inoculated milk [20] (Fig. 1). The apparatus was set to varying incubation temperatures of 39 °C, 41 °C and 43 °C for the incubation process. The yoghurt samples incubation were terminated when the pH reaches 4.6 and the samples were transferred to a refrigerator (Model: HRF-95EX Haier Thermocool, Nigeria) where the temperature of the yoghurt was allowed to reach 4 °C before taken for analysis.

Table 1 Taguchi experimental design parameters and levels

Process parameters	Code	Unit	Level 1	Level 2	Level 3
Pasteurization temperature (°C)	A	°C	80	90	100
Incubation temperature (°C)	B	°C	39	41	43
Soursop puree to sugar ratio (%)	C	%	0	50	75

Evaluation of the acidification kinetics of yoghurt

The acidification kinetics of the yoghurt samples were determined by evaluating their maximum acidification rates (MAR), time required to achieve maximum acidification and time (h) at which pH of 4.6 was reached. The maximum

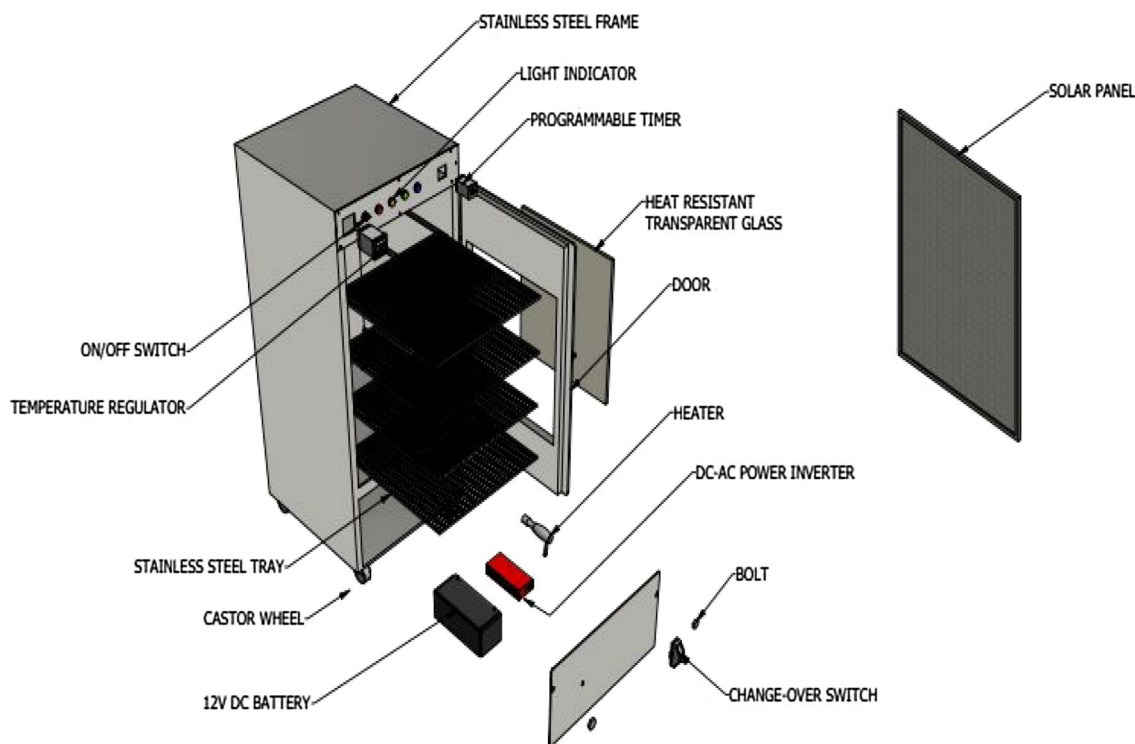


Fig. 1 Apparatus for conditioning dough and inoculated milk [20]

acidification rate is the ratio of change in pH against time, expressed in absolute values (m unit pH/min) and was determined using Eq. 1.

$$MAR = \left(\frac{dpH}{dt} \right)_{max} \quad (1)$$

pH measurement

Digital hand held pH meter (Model: CH0892, UK) was used to determine pH changes during fermentation and was monitored every 30 min by immersing the electrode into a known mass sample of yoghurt samples under incubation.

Proximate determination

The proximate composition of the yoghurt samples was analysed using the AOAC [21] methods. For moisture content, method 925.09 was used, crude fat (Method 969.24), protein (Method 950.48, Nx6.25), and ash content (Method 923.03). The crude fibre was estimated according to ISO [22]. A portion of the yoghurt sample was taken to represent the whole yoghurt. The analysis was carried out in triplicate.

Mineral content determination

Phosphorous (P), Calcium (Ca) and Potassium (K) in samples were determined by flame Atomic Absorption Spectrophotometer (VARIAN model AA240FS, United States) [23].

Determination of vitamin C content

Standard ascorbic acid solution was used to standardize the titrant 2, 6-dichlorophenol indophenol through titration. 5 ml each of the yoghurt samples were taken with a standard micro pipette into a 100 ml conical flask. The burette was filled with 0.1% dichlorophenol indophenol and adjusted to zero mark. The dye was then added drop wise into the 5 ml yoghurt sample. The continuous addition of the dye was carefully done until the blue colour remained permanently and volume of dichlorophenol indophenol used was noted. The procedure was replicated for second and third titrations noting the volume of dichlorophenol indophenol used each time [24].

Determination of total phenolic content

The total phenolic content was determined using the Folin–Ciocalteu's phenol reagent with some modifications [25]. Briefly, 1 mL of water-soluble supernatant of sample extracts was transferred into a test tube and mixed with 1 mL of 95% ethanol and 5 mL of distilled water. To each sample,

0.5 mL of 50% (v/v) Folin–Ciocalteu reagent was added and mixed. After 5 min, 1 mL of 5% Na₂CO₃ was added to the reaction mixture and allowed to stand for 60 min. The absorbance was read at 725 nm. The absorbance values were converted to total phenolics and were expressed in microgram equivalents of gallic acid per millilitre of the sample. The results were expressed as micrograms of gallic acid equivalent (GAE) per millilitre, standard curves were established using various concentrations of gallic acid in water.

Sensory evaluation of the yoghurt

Sensory evaluation was conducted to determine consumer's response and for discerning the best type of yoghurt. This was done on the nine samples of the yoghurt that had been stored for 24 h at 4 °C for proper flavour development. Sensory attributes of the samples were evaluated by 15 semi-trained panellists. The 15 panellists (10 males and 5 females, with age range of 22 to 40) were recruited from staff and final year students of department of Food Engineering and where selected based on awareness with a taste for healthy products consumption and are habitual yoghurt consumers. A 5 h round table discussions session was held to train the selected panellists on how to use nine points hedonic scale and also to select the sensory attributes that were evaluated. The nine point hedonic scale that was used ranges from “9” which is extremely like and “1” that represents extremely dislike. The colour, taste, flavour, texture and overall acceptability were the sensory attributes that were selected. After the training, the semi-trained panellists were presented with 30 ml of each of the 9 yoghurt samples in individual booths. The yoghurt samples were served in plastic disposable cups at room temperature (27 °C) and in replicate. Water was given to the panellists for palate cleansing between samples.

Statistical analysis

The data were analyzed using SPSS (Statistical Package for Social Science). The mean and standard deviation was determined using Duncan's new multiple range test of Analysis of Variance (ANOVA) at $p \leq 0.05$.

Results and discussion

Acidification profile of the yoghurt

Figure 2 revealed that the acidification profile of the nine samples of yoghurt produced slightly differs. The acidification of the yoghurt samples had four phases. The first phase was the stationary which occurred in the first 90 min of incubation of the samples where the pH practically remained unchanged, except for B and E, where the pH of the samples

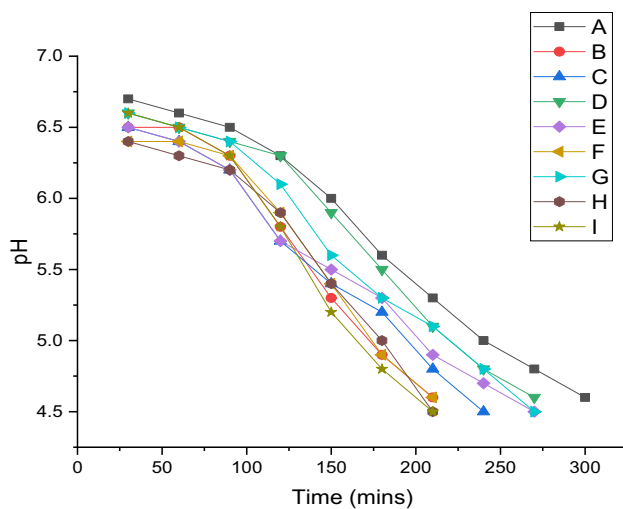


Fig. 2 Acidification profile of the nine samples of yoghurt produced

declined slowly. The differences that occur at this phase implied that the lactic acid bacteria (LAB) were adapting to the substrate due to presence of soursop puree and the incubation temperature (41 °C and 43 °C) in sample B and E. From the first 90 min up to approximately 120 min of incubation, an initial pH reduction was found virtually in all the yoghurt samples. This could be as a result of the actions of lactic acid bacteria that metabolized lactose more quickly, which also prepared the medium for the next phase, which was between 120 and 210 min where the pH fell quickly. This quick drop in pH indicated the peak activity of lactic acid bacteria. According to Bezerra et al. [26], flocculation phase coincides with peak viscosity variation in the product. In the last 60 min, pH continues to decrease but this was at a lower rate until it reached the required pH of 4.6 before it was terminated. Generally, a stationary or slight decrease in pH was shown in the first phase is dependent on the milk and soursop puree to sugar composition and incubation

temperature. This was followed by a second phase where the values of the pH decreased faster than the first phase and then third phase came with a steep curve inclination due to high pH drop. Finally, the fourth acidification phase had a little impact in the pH level and this phase was characterized by a tendency to stabilize the product. This behaviour is in agreement with Medeiros et al. [3] findings who reported on the impact of heat treatment, incubation temperature and milk source on the yoghurt kinetic acidification.

Acidification kinetics

The acidification kinetics of the yoghurt samples is presented in Table 2. The acidification kinetics were evaluated with regards to the maximum acidification rate (V_m), time to reach V_m (T_m) and time to reach pH 4.6 (T_e), which represents the isoelectric point of milk proteins [3]. High acidification rates (V_{max}) were observed in samples B, C, E and I which could be as a result of the incubation temperature (41 °C and 43 °C) when compared to the acidification rates of samples with lower incubation temperature. This was observed at approximately 120 min of fermentation, which was in line with the findings of Bezerra et al. [26]. The increase in acidification rate might be due to the fact that lactic acid bacteria had adapted to the medium. Sample A, D and G took longer time which was above 120 min to reach the maximum acidification rate (T_m) when compared with other samples. Other samples with lower time to reach (T_m) were composite yoghurt samples containing soursop puree. Lower time observed in the composite yoghurt samples was in accordance with the findings of Mbaeyi-Nwaoha and Ezeoke [27] who established that the addition of soursop fruit to the yoghurt reduced the time to reach maximum acidification rate (T_m) which was favourable for incubation. Samples A, D and G were also observed to have higher incubation time (4.5 h above) compared to the samples containing soursop puree, this was in line with Medeiros et al. [3]

Table 2 Kinetic acidification parameters of the yoghurt samples

Yoghurt samples	Pasteurization temperature (°C)	Incubation temperature (°C)	Soursop puree to Sugar ratio (%)	Maximum acidification rate (V_{max}) ($\frac{dpH}{dt}$)	Time (h) to reach maximum acidification rate (T_m)	Time (h) to reach pH (Te) 4.6
A	80.0	39.0	0.0	0.0022	3.00	5.00
B	80.0	41.0	75.0	0.0042	2.00	3.50
C	80.0	43.0	50.0	0.0042	2.00	4.00
D	90.0	41.0	0.0	0.0030	2.50	4.50
E	90.0	43.0	75.0	0.0042	2.00	4.50
F	90.0	39.0	50.0	0.0033	2.00	3.50
G	100.0	43.0	0.0	0.0033	2.50	4.50
H	100.0	39.0	75.0	0.0033	2.00	3.50
I	100.0	41.0	50.0	0.0042	2.00	3.50

who revealed that an increase in the total solids content in the milk mixture might cause a decrease in time (T_m) required to reach maximum acidification. Mbaeyi-Nwaoha and Ezeoke [27] reported similar findings. The time to reach pH 4.6 (T_e) is often referred as the time to reach the end of yoghurt fermentation [28]. The longest time to reach T_e (5 h) was observed in sample A (plain yoghurt) when the lowest incubation temperature (39 °C) was used. The shortest time to reach T_e (3.5 h) was observed when samples B, F, H and I were at temperature (41 °C and 43 °C). The soursop puree could also be an influence on the reduction in T_e as samples without soursop puree were observed to have longer T_e . The reduction of incubation temperature can cause a clear decrease in the time required to reach pH 4.6, which is justified by a decrease in lactic acid metabolic activity. Kristo et al. [29] also concluded that reducing the incubation temperature could lead to an effect on the ability of the lactic bacteria to convert lactose to lactic acid. This shows that the presence of soursop puree in the milk mixture might help to improve the activities of the lactic acid bacteria in the medium and could aid faster fermentation. Samples with high proportion of soursop puree (Sample B and H) had shorter incubation time (T_e) at a temperature of 39 and 41 °C. The shorter incubation time (T_e) could also be attributed to inclusion of soursop being added to the medium, thus made it conducive for the lactic acid bacteria to metabolize.

Moisture content

Table 3 shows effect of pasteurization temperature, incubation temperature and soursop puree on the proximate composition of yoghurt. The moisture content of the yoghurt samples ranges from 69.97 ± 0.01 to $72.77 \pm 0.01\%$. Sample H (100 °C pasteurization temperature, 75% soursop puree ratio and 39 °C incubation temperature) had the lowest moisture content ($69.97 \pm 0.01\%$) while the sample G (100 °C pasteurization temperature, 0% soursop puree ratio and 43 °C incubation temperature) had the highest moisture content ($72.77 \pm 0.01\%$). It was observed that yoghurt samples (B, E and H) with 75% soursop puree ratio had the lowest moisture content. Therefore, it could be deduced that pasteurization temperature and incubation temperature did not have a significant influence on the moisture content of the yoghurt samples. However, the soursop puree to sugar ratio was observed to influence the moisture content i.e. moisture content decreased with increase in the concentration of soursop puree to sugar ratio. The moisture contents observed in this study fell within the range of moisture contents of 74.57% to 79.26% of yoghurt with graded level of soursop reported by Ekere [30]. The similarity in the range of the moisture content observed could be due to the use of soursop

Table 3 Effect of pasteurization temperature, incubation temperature and soursop puree to sugar ratio on the proximate composition

Sample	Pasteurization temperature (°C)	Incubation temperature (°C)	Soursop puree ratio (%)	Moisture content (%)	Crude protein (%)	Crude fat (%)	Ash content (%)	Carbohydrate content (%)	Crude fiber (%)
A	80	39	0	72.46 ± 0.01^a	4.05 ± 0.35^{bc}	3.01 ± 0.01^f	0.70 ± 0.01^{abc}	18.52 ± 1.90^a	0.09 ± 0.01^{ab}
B	80	41	75	70.02 ± 0.01^c	6.55 ± 0.45^a	4.00 ± 0.01^a	0.67 ± 0.02^{bc}	19.42 ± 0.02^a	0.09 ± 0.01^b
C	80	43	50	71.80 ± 1.42^{ab}	4.64 ± 0.03^b	3.14 ± 0.01^e	0.68 ± 0.01^{abc}	19.62 ± 0.50^a	0.09 ± 0.01^{ab}
D	90	41	0	71.93 ± 0.01^{ab}	4.10 ± 0.42^{bc}	2.85 ± 0.01^h	0.69 ± 0.01^{abc}	18.60 ± 0.04^a	0.10 ± 0.00^{ab}
E	90	43	75	70.05 ± 0.01^c	6.55 ± 0.50^a	3.87 ± 0.01^b	0.69 ± 0.01^{abc}	20.91 ± 0.41^b	0.11 ± 0.01^{ab}
F	90	39	50	70.86 ± 0.01^{bc}	3.99 ± 0.03^{bc}	2.97 ± 0.01^g	0.67 ± 0.01^{bc}	18.58 ± 0.05^a	0.10 ± 0.00^{ab}
G	100	43	0	72.77 ± 0.01^a	3.85 ± 0.01^c	3.20 ± 0.01^d	0.71 ± 0.01^{ab}	18.23 ± 0.35^b	0.12 ± 0.01^a
H	100	39	75	69.97 ± 0.01^c	4.25 ± 0.21^{bc}	3.88 ± 0.01^b	0.72 ± 0.02^a	19.51 ± 0.24^b	0.10 ± 0.01^{ab}
I	100	41	50	71.69 ± 0.01^{ab}	3.95 ± 0.21^{bc}	3.44 ± 0.02^c	0.65 ± 0.04^c	19.44 ± 0.25^b	0.10 ± 0.01^{ab}

Values of mean along the same column with different superscript are significantly different at $p \leq 0.05$

fruit juice in the study reported by Ekere [30] and the use of soursop puree in this study.

Crude protein

Table 3 shows that the crude protein content ranges from $3.85 \pm 0.01\%$ to $6.55 \pm 0.5\%$. Samples B (80 °C pasteurization temperature, 75% Soursop puree ratio and 41 °C incubation temperature) and sample E (90 °C pasteurization temperature, 75% Soursop puree ratio and 43 °C incubation temperature) had the highest crude protein content while sample G (100 °C pasteurization temperature, 0% Soursop puree ratio and 43 °C incubation temperature) had the lowest protein content of all the yoghurt samples. There was no significant difference in the crude protein content of sample B and E. From the result, it could be deduced that the protein content of the yoghurt was influenced by the amount of soursop puree but was not influenced by the pasteurization and incubation temperature. This could be attributed to the protein content of soursop which was reported to be 2.91 g in 100 g of soursop by Enweani et al. [31]. In addition, the protein content in the soursop puree complements the protein that is present in the skimmed milk used, thus influences the protein content of the yoghurt. Ekere. [30] reported that skimmed milk is rich in protein. The protein content observed in this study was higher than 1.95 to 2.70%, 1.29 to 3.52% and 3.4 to 5.6% that were reported by Matela et al. [32], Umeh et al. [33] and Janhoj et al. [34], respectively. The difference in the protein content could be attributed to the inclusion of soursop puree used in the yoghurt production as against plain yoghurt and yoghurt flavoured with mango juice used in other studies earlier reported.

Crude fat

The crude fat content ranges from $2.85 \pm 0.01\%$ to $4.0 \pm 0.01\%$ as shown in Table 3. The minimum crude fat content was observed in yoghurt of sample D (90 °C pasteurization temperature, 0% soursop puree ratio and 41 °C incubation temperature) which was a plain yoghurt while the maximum crude fat content was observed in sample B (80 °C pasteurization temperature, 75% soursop puree ratio and 41 °C incubation temperature). Fat content was found to gradually increase with addition of the soursop puree and the effect of the pasteurization temperature could be observed to be minimal on the crude fat content. Yoghurt sample with higher percentage of soursop puree (75%) had higher crude fat content compared to yoghurt samples with 0% soursop puree to sugar ratio. The fat content of yoghurts varies depending on the product, ranging from approximately 10% fat for full fat Greek style yogurts, 3% fat for whole milk yogurts, 1.7% fat for low fat yogurts and non-fat varieties containing less than 0.3% fat [35]. The highest fat content

of the yoghurt sample observed in this study was higher than 3.5% reported by Matela et al. [32] for plain yoghurt samples that were purchased from Maseru, the Kingdom of Lesotho. The difference in the crude fat content could be as a result of the inclusion of soursop puree in the yoghurt samples.

Ash content

Table 3 shows that the ash content ranges from $0.65 \pm 0.04\%$ to $0.72 \pm 0.02\%$. Sample H (100 °C pasteurization temperature, 75% soursop puree ratio and 39 °C incubation temperature) had the highest ash content while sample I (100 °C pasteurization temperature, 50% soursop puree ratio and 41 °C incubation temperature) had the lowest ash content. There were no significant differences in the ash content of most of the yoghurt samples. The low ash content observed in the yoghurt samples might be due to the low level of ash in the soursop puree. From the results, it can be deduced that the ash content was influenced by the incubation temperature and pasteurization temperature. Ash is the residue remaining after water and organic matter have been removed by heating in the presence of oxidizing agents [25]. The range for the ash content of the yoghurt samples in this study were within the same range of 0.28–0.95% reported by Matela et al. [32] for plain yoghurt samples. The similarities observed in the ash content ranges further clarifies that soursop puree does not have much influence on the ash content of the yoghurt sample.

Carbohydrate content

Carbohydrate is the major constituent of milk that is converted to lactic acid during yoghurt production [36]. Carbohydrate content ranged from $18.23 \pm 0.35\%$ to $20.91 \pm 0.41\%$ in Table 3. The sample with the highest carbohydrate content was sample E (90 °C pasteurization temperature, 75% soursop puree ratio and 43 °C incubation temperature) while the yoghurt sample with the lowest moisture content was sample G (100 °C pasteurization temperature, 0% soursop puree ratio and 43 °C incubation temperature). Yoghurt samples with high soursop puree to sugar ratio were observed to have high carbohydrate content. Soursop is a fruit rich in sugar which might compliment the carbohydrate content of yoghurt. Bradie and Schauss. [32] reported that soursop had a high carbohydrate content of 14.63 g in 100 g. The range observed for carbohydrate contents in this study were within the ranges of 9.41–19.33% and 13.66–19.20% reported by Matela et al. [32] and Ekere.[30].

Fibre content

Table 3 revealed that the fibre content ranges from $0.09 \pm 0.01\%$ to $0.12 \pm 0.01\%$. Yoghurt samples of A, B and

C had the lowest fibre content ($0.09 \pm 0.01\%$), while sample G had the highest fibre content ($0.12 \pm 0.01\%$) of all yoghurt samples. According to Bradie and Schauss [37], soursop as a low fibre content of 0.79 g in 100 g. This could be the reason why there was no significant difference in the fibre content between plain yoghurt and yoghurt incorporated with soursop. This result is in agreement with the observation of authors who stated that yoghurts usually have poor fibre content level because they are milk and water based products. Therefore, considerable increase in the fibre content of soursop yoghurt samples was as a result of the addition of soursop puree. The result obtained for the range of fibre content of the yoghurt samples was lower than 1.26–2.87% reported by Umeh et al. [33] for African bush mango yoghurt samples and higher than 0.01–0.08% reported by Matela et al. [32] for plain yoghurt.

Vitamin C content

Figure 3 shows the effect of the pasteurization temperature, the incubation temperature and the soursop puree to sugar ratio on the vitamin C content of the yoghurt samples. The vitamin C content ranged between 36.10 and 72.76 mg/l. The highest amount of vitamin C content was observed in sample E at 90 °C pasteurization temperature, 75% Soursop puree ratio and 43 °C incubation temperature, while the lowest vitamin C content was observed in sample G at 100 °C pasteurization temperature, 0% Soursop puree ratio and 43 °C incubation temperature. The difference in the vitamin C content of sample E and sample G could be as a result of the high presence of soursop puree in sample G which might contribute to the high level of vitamin C present in the yoghurt. According to Bradie and Schauss [37], soursop has high vitamin C content of

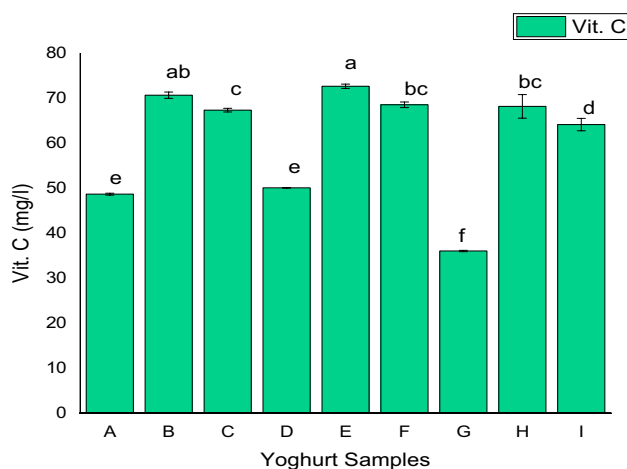


Fig. 3 Effect of the pasteurization temperature, incubation temperature and soursop puree to sugar ratio on the vitamin C content of yoghurt sample

29.6 mg in 100 g. Soursop is a kind of fruit rich in antioxidants such as vitamin C which helps to destroy free molecular radicals [38]. The level of inclusion of soursop puree in the samples influenced the vitamin C contents of the final products. This result corroborated with the findings of Ekere [30], who revealed that inclusion of soursop juice in yogurt increased its vitamin C content.

Phosphorus

Figure 4 shows the effect of the pasteurization temperature, incubation temperature and soursop puree to sugar ratio on the phosphorus content of yoghurt samples. The phosphorus content varied from 120 to 519 mg/l. Sample A (80 °C pasteurization temperature, 0% soursop puree ratio and 39 °C incubation temperature) had the lowest phosphorus content while sample H (100 °C pasteurization temperature, 75% Soursop puree ratio and 39 °C incubation temperature) had the highest phosphorus content. There were no significant differences in phosphorus content of the plain yoghurt in samples A, D and G as shown in Fig. 4. Out of all the parameters used in the production of the yoghurt sample, samples with high amount of soursop puree to sugar ratio was observed to have the most influence on the phosphorus content of the yoghurt samples. The phosphorus content increased with an increase in the percentage of the soursop puree added to the yoghurt. This increase could be as a result of the high phosphorous content of 27.7 mg per 100 g present in soursop as reported by Bradie and Schauss [37]. The results obtained in this study fell within the range of 362.27–397 mg/kg reported by Kibui et al. [39] on yoghurt samples enriched with chia seeds, suggesting that both soursop and chia seed might be rich in phosphorus.

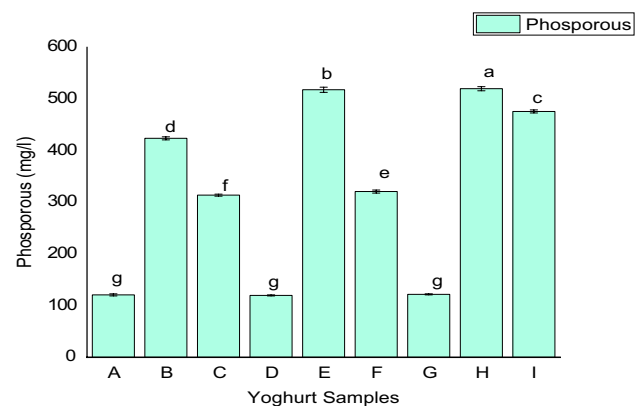


Fig. 4 Effect of the pasteurization temperature, incubation temperature and soursop puree to sugar ratio on the phosphorus content of yoghurt samples

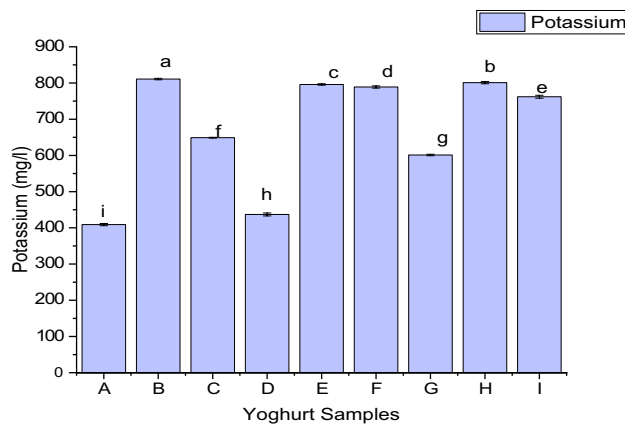


Fig. 5 Effect of the pasteurization temperature, incubation temperature and soursop puree ratio on the potassium content of yoghurt sample

Potassium content

Figure 5 shows the effect of pasteurization temperature, incubation temperature and soursop puree ratio on the potassium content of yoghurt samples. The potassium content ranged from 408.5 to 810.5 mg/l. The highest amount of potassium content was observed in sample B (80 °C pasteurization temperature, 75% soursop puree ratio and 41 °C incubation temperature) and the least value was observed in sample A (80 °C pasteurization temperature, 0% Soursop puree ratio and 39 °C incubation temperature). There were significant differences $p \leq 0.05$ in the potassium contents of all the yoghurt samples, this implies that there were no similarities in the potassium content of each sample of yoghurt produced. The sample with the least value of potassium was a plain yoghurt sample, while that with the highest value was observed to be the yoghurt sample containing high percentage of soursop puree to sugar ratio. The trend in Fig. 5 also shows that the potassium content increased with an increase in the soursop puree. Soursop has a good amount of potassium content which supplements the potassium content of skimmed milk. Therefore, addition of soursop puree would yield an increase in the potassium content of yoghurt.

Calcium content

Figure 6 shows that the effect of the pasteurization temperature, incubation temperature and soursop puree to sugar ratio on the calcium (Ca) content of the yoghurt samples. The calcium content ranged from 298.5 mg/l in sample A (80 °C pasteurization temperature, 0% soursop puree ratio and 39 °C incubation temperature) to 623.5 mg/l in Sample B (80 °C pasteurization temperature, 75% soursop puree ratio and 41 °C incubation temperature). This result shows that yoghurt produced with soursop puree had the highest

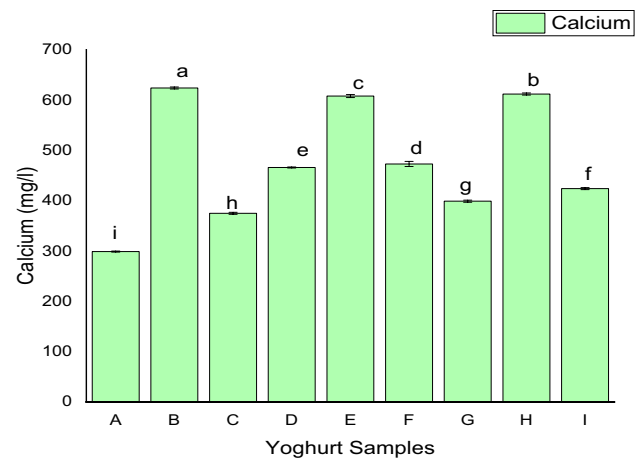


Fig. 6 Effect of the pasteurization temperature, incubation temperature and soursop puree ratio on the calcium content of yoghurt sample

calcium content while plain yoghurt samples had the lowest calcium content. Furthermore, the soursop puree could be deduced to have the most effect on the calcium content of the yoghurt. There were significant differences ($p \leq 0.05$) in the calcium content of all the yoghurt samples, this difference could be attributed to the different processing parameters used during production. The higher level of calcium in the samples containing soursop puree could also be attributed to the addition of soursop to the milk which increases the calcium content. Plain yoghurt contains 120.8 mg of calcium per 100 g [38], whereas soursop has a calcium content of 10.3 mg per 100 g [37]. The results for the calcium content of the yoghurt samples were higher than 5.2–8.0 mg per 100 g reported by Ekere [30] on graded yoghurt flavoured with soursop juice. The changes observed could be due to difference in processing conditions, as Ekere [30] used soursop juice while this study used soursop puree.

Total phenolic content of the yoghurt

The effect of pasteurization temperature, incubation temperature and soursop puree to sugar ratio on the phenolic content of the yoghurt samples is shown in Fig. 7. The total phenol content ranged from 1.07 to 1.44 mg GAE/L. The highest amount of phenolic content was observed at sample H (100 °C pasteurization temperature, 75% soursop puree ratio and 39 °C incubation temperature) and the least value was observed in sample A (80 °C pasteurization temperature, 0% soursop puree ratio and 39 °C incubation temperature). The plain yoghurt samples with no soursop puree to sugar ratio was observed to have low phenolic content. However, it was noted that the addition of various levels of soursop puree to sugar ratio in the yoghurt samples caused an increase in total phenolic content of the yoghurt samples. Phenolic compounds are products of secondary metabolism

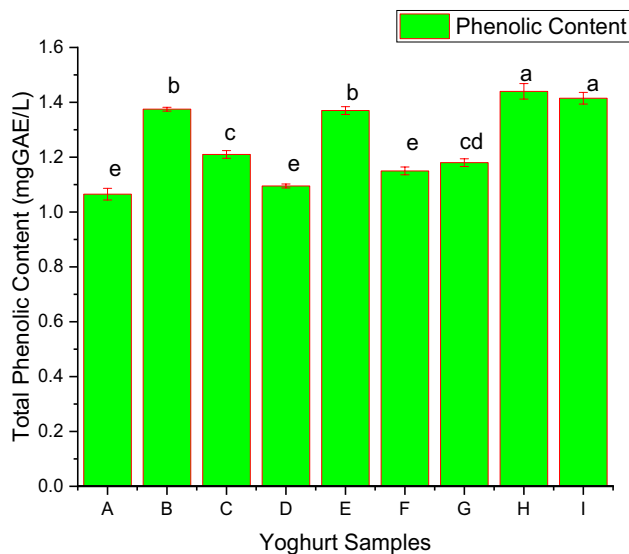


Fig. 7 Effect of the pasteurization temperature, incubation temperature and soursop puree ratio on the phenolic content of yoghurt sample

in plants and are good sources of natural antioxidants in human diets [35]. This increase was observed because soursop contained a lot of antioxidants such as phenol [40]. Senadeera et al. [12] reported an increase in total phenolic content of probiotic yoghurt incorporated with various *Annona species* pulp. The similar trend in the results could be as a result of inclusion of soursop (*Annona species*) in yoghurt.

Sensory attributes of the yoghurt

The effect of pasteurization temperature, incubation temperature and soursop puree to sugar ratio on the sensory attributes of yoghurt sample is presented in Table 4. The mean scores for the yoghurt colours range from 6.20 ± 1.26 to 8.27 ± 0.70 . Sample I with 75% soursop puree to sugar ratio had the lowest mean score of all the

samples while sample A with 0% soursop puree to sugar ratio had the highest colour score. Plain yoghurt samples with 0% soursop had the highest score for colour. There were no significant differences ($p \geq 0.05$) in the colour of samples C, D, E, F, and G and this could be due to the presence of soursop puree in the yoghurt samples. The brownish white colour of the soursop mesocarp might be responsible for the low scores observed. The mean scores for taste ranged from 3.80 ± 1.94 to 7.73 ± 0.96 . Sample I had the lowest score for taste while sample A had the highest mean score. The sample with the highest score for taste was plain yoghurt. The taste of the soursop puree can be a major reason for the low score obtained as soursop pulp has a sweet-sour taste [12]. The flavour of the yoghurt samples followed the same trend as the taste. The preference for the flavour of the products reduced with increasing level of soursop puree addition. The score for flavour ranges from 3.47 ± 2.07 to 7.67 ± 0.98 . The scores for flavour were observed to be lowest in sample I and highest in sample A. The mean score for texture ranges from 3.93 ± 1.83 to 7.33 ± 1.05 . Sample I gave the lowest texture value while the highest was observed in sample A. The overall acceptability of the yoghurt samples ranges from 3.87 ± 1.96 to 8.00 ± 0.85 . There was a reduction in the overall acceptance of the yoghurt samples as the percentage of soursop puree to sugar ratio increases. High mean values (> 6.5) was observed in yoghurt of sample B with 75% soursop puree to sugar ratio. Generally, the mean values for overall acceptability of most yoghurts samples with the inclusion of soursop were more than 'slightly like' (5.0) except for samples E, H and I. In the overall sensory attributes, plain yoghurt scored higher than soursop flavoured yoghurts. This might be due to the fact that the panellists were more accustomed to plain yoghurt rather than the soursop flavoured yoghurts. This could also be attributed to the sweet-sour taste accustomed to soursop fruits.

Table 4 Effect of pasteurization temperature, incubation temperature and soursop puree to sugar ratio on the sensory attributes of yoghurt

Samples	Colour	Taste	Flavour	Texture	Overall acceptability
A	8.27 ± 0.70^a	7.73 ± 0.96^a	7.67 ± 0.98^a	7.33 ± 1.05^a	8.00 ± 0.85^a
B	7.53 ± 1.12^{ab}	5.80 ± 1.42^{bc}	6.00 ± 1.10^{bc}	6.67 ± 1.35^{ab}	6.80 ± 1.08^{ab}
C	6.93 ± 2.09^{bc}	3.93 ± 2.34^{de}	5.27 ± 1.87^{bcd}	6.20 ± 2.24^{ab}	5.60 ± 1.64^{bcd}
D	6.5333 ± 2.0^{bc}	5.47 ± 2.29^{cd}	5.27 ± 2.37^{bcd}	4.40 ± 2.75^{cd}	5.13 ± 1.99^{cde}
E	6.47 ± 1.64^{bc}	4.73 ± 2.15^{cde}	4.87 ± 1.96^{cde}	4.20 ± 2.04^{cd}	4.47 ± 2.62^{de}
F	7.00 ± 1.6^{bc}	4.53 ± 2.20^{cde}	5.07 ± 1.7^{bcd}	5.13 ± 2.26^{bcd}	5.20 ± 1.94^{cde}
G	6.80 ± 1.32^{bc}	7.13 ± 1.13^{ab}	6.47 ± 1.25^{ab}	5.67 ± 1.84^{bc}	6.60 ± 1.64^{abc}
H	7.13 ± 1.64^{abc}	4.80 ± 2.68^{cde}	4.40 ± 2.56^{de}	4.53 ± 2.30^{cd}	4.67 ± 2.44^{de}
I	6.20 ± 1.26^c	3.80 ± 1.94^e	3.47 ± 2.07^e	3.93 ± 1.83^c	3.87 ± 1.96^e

Values are mean $\pm$ standard deviation. Means on the same column with different superscripts are significantly different ($P \leq 0.05$)

Conclusions

Yoghurt produced with the substitution of sugar with soursop puree improves the acidification kinetics, proximate composition, vitamin C, mineral contents and total phenolic content, respectively. Maximum acidification rate were achieved in yoghurt samples that have 50% and 75% of soursop inclusion during yoghurt production than the plain yoghurt samples (0% soursop to sugar ratio) that does not have soursop puree. The soursop puree to sugar ratio has an influence on the moisture content of the yoghurt. Yoghurt samples which were produced with different ratios of soursop puree to sugar were higher in protein content, fat content and carbohydrate content as against that of plain yoghurt samples. Pasteurization and incubation temperature was influential in determining the ash content of the yoghurt. The yoghurt samples produced with soursop puree had high vitamin C as compared with plain yoghurt and it increases with increasing ratio of soursop puree to sugar ratio. The inclusion of soursop puree influences the mineral content such as the calcium, potassium and the phosphorus content of the yoghurt at different incubation and pasteurization temperatures. The total phenolic content of the yoghurt samples were seen to have increased due to the inclusion of soursop puree. Yoghurt with 75% soursop to sugar ratio was the most preferred of all yoghurt samples with soursop based on the sensory attributes. This study showed that yoghurt with varying soursop to sugar ratio could be produced without having negative impact on the consumer acceptability. The outcome of this study could help the yoghurt industry develop new yoghurt varieties.

Declarations

Conflict of interest The authors declare that they have no conflict of interest.

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