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Full Length Research

Production and quality evaluation of soy-tigernut milk

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ABSTRACT: This study was conducted to determine the proximate, minerals, vitamins, functional properties, phytochemicals and microbial growth of soy-tiger nut milk. Dried tiger nut tubers and soybean seeds were sorted, washed, soaked, dehulled (soybeans), milled and filtered to obtain fresh milk. The results obtained showed that there was a significant difference ($p<0.05$) in the proximate composition of samples. The ash, moisture, fibre, protein, and carbohydrate content ranged from 1.30% - 2.93%, 73.00% - 81.33%, 0.02% - 1.83%, 2.43% - 3.93%, 5.06% - 8.48%, 6.34% - 16.93%, respectively with PAP having high carbohydrate content. The mineral composition comprises calcium, magnesium, iron and phosphorous content with FAD having the highest calcium and phosphorous content. The vitamin composition includes vitamins A, B_2, C and B_6. The functional composition comprises of water absorption capacity and solubility index with PAP having the highest water absorption capacity. The phytochemicals composition ranges from 6.23 - 8.50 and 11.60- 12.96 mg/100g for flavonoids and phenols and ZAK has the highest phenols. In microbial load, the bacteria organisms isolated were *Bacillus spp*, *Micrococcus spp, Klebsiella spp, Staphylococcus spp, Corynebacterium spp* and *Pseudomonas spp* and the fungi isolated were *Candida spp, Fusarium spp, Penicillium and Rhizopus spp* and *Saccharomyces cerevisiea*.

Keywords: Functional foods, imitation milk, probiotic effect, soy milk, tigernut milk.

INTRODUCTION

Milk is a whitish liquid containing proteins, fats, lactose, vitamins and minerals, secreted by the mammary glands of all adult female mammals after childbirth. It serves as food for their young immediately after birth (Encyclopaedia Britannica, 2024). According to Antunes *et al.* (2022), milk is a complete food, containing all the necessary ingredients such as very good proteins and essential micronutrients, including vitamins and minerals. It should be noted that milk culture is infused with the sanctity of the cow in ancient Egypt, Iran and India. In Europe, the monks, including the Benedictines in the Middle Ages were the main producers of cheese, for example, Bishop and Munster. Milk is a liquid substance and an excellent source of all nutrients except iron and ascorbate.

Milk has been identified to contain both the micro and macronutrients required for the growth and development of the infant; with high biological value presenting all essential amino acids and possessing high digestibility (Gaucheron, 2011; Muehlhoff *et al.,* 2013). Studies reveal that milk and dairy products could provide protective action against prevalent chronic diseases with minimal or no adverse effects (Antunes *et al.,* 2022; Thorning *et al.,* 2016). The vital protective function offered by milk has led to increased research necessitated by the intolerance to milk lactose and allergenic reactions propelling the need for choices (Jeske *et al.,* 2017; Jeske *et al.,* 2018). Plant-based alternative milk consumption is on the increase which has led to a sharp rise in the market share of the product, as a healthy alternative as it possesses less saturated fats, and the absence of cholesterol with a peak in non-saturated fats but coconut milk (Thorning *et al.,* 2016; Jeske *et al.,* 2017; Jeske *et al.,* 2018; Bridges, 2018;

Röös *et al.,* 2018).

Different varieties of milk include whole milk, skimmed milk, flavoured milk, evaporated milk, sweetened condensed milk, dry whole milk and many more (Turler-Inderbitzin, 2012). Today, the meaning of milk gives more consideration to its constituent substances than its source or origin owing to the discovery of soy and other milk of plant origin at the turn of the 20th century. These plant milks are highly nutritious, beneficial to health and cheap (Vucenik and Shamsuddin, 2003; Sudheer *et al.,* 2004; Sacks *et al.,* 2006). In some developing countries, the cost of dairy milk and their product are prohibitive. Also, the inability of some individuals to consume dairy milk containing lactose led to the discovery of “Imitation or faux milk”.

Imitation milk is a non-dairy product manufactured to resemble milk and can be used as a substitute for milk. Basically, it contains water, vegetable fat, corn sugar, starch, vegetable protein, sodium, caseinate, stabilizers, vitamins, and minerals may also be added to it. They are primarily obtained from plant sources such as coconut, tiger nut, soybean, almond, rice, oat, peanut, etc. They contain no cholesterol and are lactose free which makes them more suitable for lactose intolerant individuals. It is worth noting that milk from plant sources is seen as a radiating hope as well as an ally in fighting against malnutrition and hunger (Belewu *et al.,* 2007). Milk-like beverages manufactured from oilseed preparation have great potential as nutritional substitutes, especially in cultures where cow milk is insufficient, too expensive or indigestible (Isanga and Zhang, 2009).

Nevertheless, the use of plant-based milk alternatives is of concern in nutritional and health studies arising from increased disposing of the body to diseased conditions in infants and children. Antunes *et al.* (2022) reported the positive effect of plant-based milk alternatives on obesity, type II diabetes and Alzheimer's diseases, while there had been reported cases of its negative effects on prostate cancer and Parkinson's diseases.

Besides macronutrients, milk also contains numerous nutrients (micro and macro) such as calcium, selenium, riboflavin, vitamin B_{12} and pantothenic acid (vitamin B_5) which significantly contribute towards the overall growth and maintenance of the body system (Makinen *et al.,* 2016). Though milk is considered to be a complete food, yet limited availability or near absence of certain minerals such as iron, folate, and some other biomolecules (amino acids) restricts its recommendation as a complete food for infants older than 12 months (Vanga *et al.,* 2015). Further, lactose intolerance is a continuously growing disease throughout the developed world, especially in the old age population groups restricting the use of milk and milk products, especially by the elder population. Additionally, the limited availability of milk in some specific geographic locations (arid regions), high price and the presence of some potent pathogens (*Salmonella spp.* and *Escherichia coli* O157:H7) which can cause disease outbreaks and some specific health problems (Vanga and Raghavan, 2018) like cholesterol, cow’s milk allergy mainly seen among some infants and children, antibiotic residues, vegetarianism and vegan diets may be assumed as the possible reason for conceptualization and development of milk-alternatives or milk-substitutes or milk-analogs. Expectations of present consumers for more healthy and palatable food choices have driven the dairy industry to expand their knowledge beyond conventional beverages with health benefits equating milk with consumption at recommended levels (Grant and Hicks, 2018). Medical complications (lactose intolerance, allergens, negative association of bovine milk fat with cardiovascular disease) with conventional milk or some lifestyle-related issues (high protein requirement, balanced amino acid profile, probiotic beverages) have resulted in consumers more interested in vegan diets over normal mammalian milk. Though the market for milk analogs is governed by specific choices and influenced by various opinions developed over time, yet the preference for milk analogs is not out of necessity but out of the search for cheaper and more efficient food sources. The consumption of plant-based nondairy beverages has been reported in early civilizations all over the world with limited availability to the local market, preparation at a traditional gathering or home scale (Röös *et al.,* 2018).

Soybean (*glycine max*) is one of the most important oil crops in the world which has great importance as a legume. It is native to East Asia and widely grown for its edible beans for numerous uses. It belongs to the family Leguminosae sub-family Papilionaceae. It contains a significant amount of phytic acid, dietary minerals and vitamin B. Consumption of soybeans helps reduce the risk of heart diseases, and can be used as a substitute for people that are allergic to animal milk. Soybean is known for their high food value for centuries and it was used for food purposes (soymilk, tofu, douche, hamanatto, miso shoyu, doufu, tempeh, soy flour, green beans, roasted soynuts, soybeans sprouts, soy sauce, soy meal, soy vegetable oil, fermented beans paste etc.), with the dawn of civilization (Norberg and Deutsch, 2023).

The soybean plant (*Glycine max*) belongs to the good complement to cereal protein, which is low in the legume family. On average, dry soybean contains roughly 40% protein, 20% oil, 35% soluble (sucrose, raffinose, starchyose etc.) and insoluble dietary carbohydrate and 5% ash. Fresh soybean has approximately 14% moisture (Liu, 2004). Humans can easily digest soy protein. About 92-100% of soy protein is digestible in humans (Riaz, 1999).

Glycine max, with 40% protein and 20% fat assumes the most predominant position in solving the nutritional imbalance prevailing. It not only provides quality macronutrients but also various other micronutrients, which are otherwise required to fight against malnutrition.

As a rich protein source, soybean can furnish protein supply to bridge up the deficiency gap at low-cost than any other crop (Rehman *et al.* 2007). Among the numerous soy food items, soymilk (extract of soybeans) was the first product ever prepared and consumed by humans since long ago. Soymilk not only provides protein but also is a source of carbohydrates, lipids, vitamins and minerals. Soymilk is a healthy drink and is important for people who are allergic to cow milk protein and lactose (Rehman *et al.,* 2007).

Tiger nut (*Cyperus esculentus*) is a perennial emergent grass-like plant with spheroid tubers, pale yellow cream kernel surrounded by a fibrous sheath. It is known as yellow nut sedge, earth or ground almonds, “souchet” in French, “ermandeln” in German and “chufa” in Spanish (TTSL, 2005). It is an underutilized tuber belonging to the family of *Cyperaceae*. It grows freely and is consumed widely in Nigeria, other parts of West Africa, East Africa, parts of Europe particularly Spain as well as the Arabian Peninsula. Tiger nut grows in the wild, along rivers and is cultivated on a small scale by rural farmers mostly in the Northern states of Nigeria and is locally called “aya” in Hausa; “aki awusa” in Igbo; “ofio” in Yoruba, and “isipaccara” in Efik. *C. Esculentus* is a tuber rich in energy content (starch, fat, sugar, and protein), minerals (mainly phosphorous and potassium) and vitamins E and C (Moore, 2004).

Tiger nut has a lot of health and economic benefits; it aids in the prevention of heart diseases, thrombosis, reduces the risk of colon cancer, and possesses good therapeutic quality (Farre, 2003; Bixquert, 2003; Yu *et al.,* 2022; Ejoh and Djomdi, 2006). Moore (2004) stated that “the expansion of tiger nut milky drinks would significantly help the research linking tiger nut milk to healthier cholesterol levels and non-dairy manufacturers. Yu *et al.* (2022) noted that tiger nut has the potential to scavenge free radicals, inhibit *in-vitro* lipid peroxidation, depress inflammation, and possess medicinal applications in anti-apoptosis. This could also gain a boost from an increased consumer interest in healthier foods. This tuber is also suitable for diabetics and for those with intent on losing weight. There are a variety of products that can be produced from tiger nut though there is very little documentation at large. Various food processing techniques can be applied to tiger nut processing to modify its appearance, develop its natural flavour, stimulate the digestive juices, add variety to the menu, make it easily digestible and bio-available, destroy harmful microorganisms, improve its nutritional value and prevent decomposition. They could include, tiger nut coffee (which is produced when tiger nut tubers are roasted and blended to give that coffee colour and taste), tiger nut wine (which can be produced when the tiger nut milk is allowed to ferment), tiger nut milk etc. During the era when ancient Mesopotamia was between the Rivers Tigris and the Euphrates, tiger nut was classified as a medicinal drink due to its being highly energetic and diuretic, rich in minerals predominantly potassium and phosphorous and also vitamin C and E (AOAC, 2023).

For the purpose of this research, possible concentrations of tiger nut and soy milk were formulated with the intention of evaluating the physicochemical and microbiological quality of the product as a healthy alternative to cow's milk.

MATERIALS AND METHODS

Source of raw materials

The dried tiger nut tubers and soybean grains were purchased from Eke Awka Market, Awka, Anambra State, Nigeria. The equipment and chemicals were of analytical grade.

Experimental design

The experiment was designed using Mixture Design (Minitab Version 21). The samples were randomly selected from a batch of population and were tested for nutritional composition.

Production of soy-tiger nut milk

Tiger nut milk production

One (1) kg of tiger nut tuber was sorted, washed, and soaked for 48 hours in 700 ml of water. The tuber was drained, properly rinsed and milled with potable water, the slurry was filtered using a clean muslin cloth and the milk extract was heated for 5 seconds at 72°C after which it is hot filled into a sterilized bottle and allowed to cool as shown in Figure 1.

Production of soymilk from soybean

One (1) kg of soybean was sorted, washed, and soaked for 18 hours in 3 L of potable water in a ratio of 1:3. The soybean was properly drained and dehulled by rubbing the beans with the palm. It was milled using a Binatone auto-clean blender and the resulting slurry was filtered using a clean muslin cloth and the milk extract was pasteurized for 15 minutes at 72°C. It was hot-filled into a sterilized bottle and allowed to cool (Figure 2).

Production of soy-tiger nut milk

The processing of tiger nut milk is shown in Figure 3. Tiger

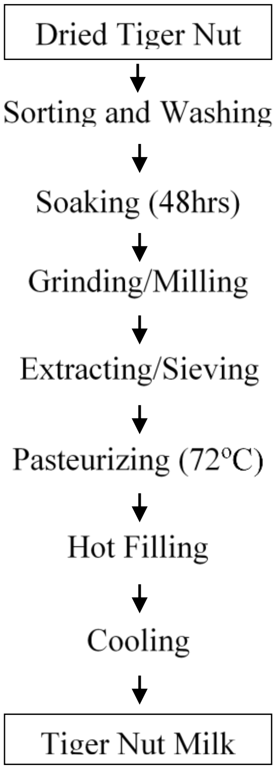

Figure 1. Flow chart for the production of tiger nut milk drink.

nut milk and soymilk were mixed at different proportions to obtain the final product. This was done using an electric blender operated at full speed for 10 minutes. The resulting blend was also homogenized and pasteurized at 72°C for 5 seconds. It was hot-filled and cooled immediately to room temperature.

Methods of analysis

Proximate composition of soy-tiger nut milk

The proximate compositions such as fiber, protein, fat, ash and moisture contents were analyzed using AOAC (2023) methods. The carbohydrate content was obtained by difference.

Determination of microbial quality

The microbial counts were enumerated using AOAC (2023) methods.

Determination of vitamins

The vitamins such as vitamins A, B_2, C and vitamin B_6 were determined by the AOAC (2023) Method.

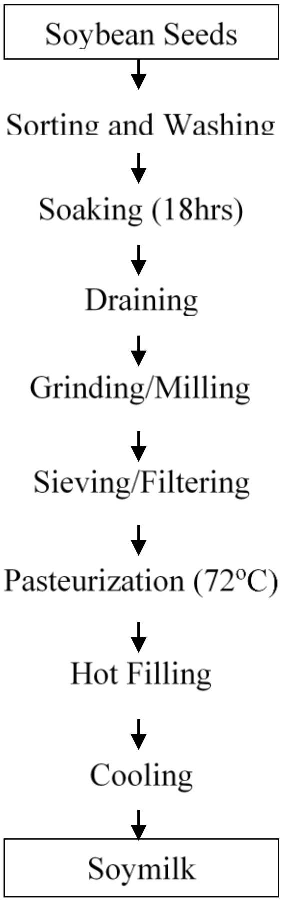

Figure 2. Flow chart for the production of soymilk.

Determination of functional properties

The water absorption capacity and solubility index of the soy-tigernut samples were determined according to the method adopted by Onwuka (2018).

Mineral determination

The mineral content (calcium, magnesium, phosphorous, iron, and manganese) was determined according to the standard methods of the AOAC (2023) method.

Sensory evaluation

A semi-trained panel of 20 judges made up of males and females was used. The panellists evaluate the product based on aroma, appearance, texture, colour, taste, and overall acceptability using a 9-hedonic scale, where 9 will be equivalent to like extremely and 1 equivalent to dislike extremely.

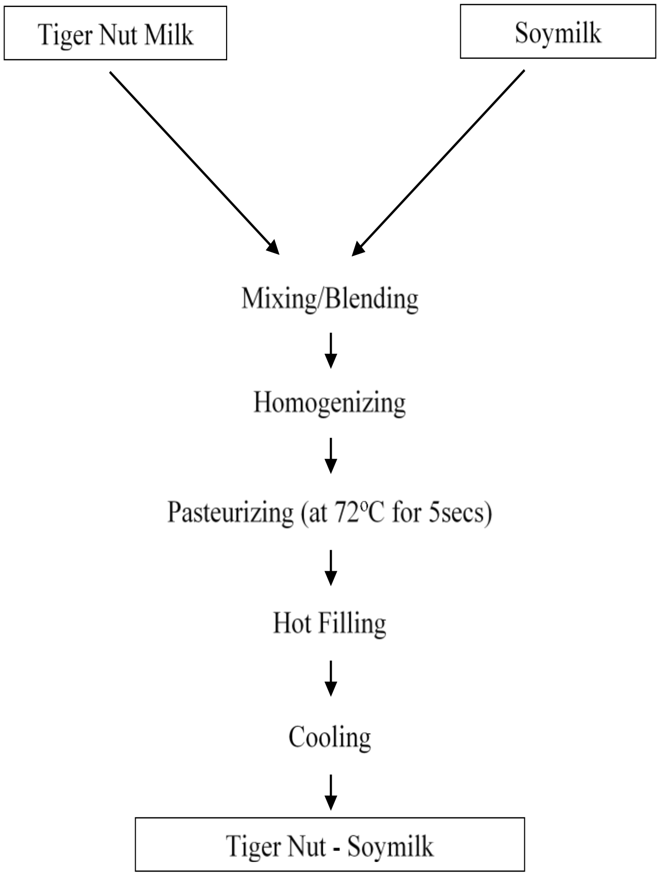

Figure 3. Flow chart for the production of Soy-tigernut milk.

Statistical analyses

The mean and standard deviation of the result data from the experimental was calculated and analyzed using Analysis of Variance (ANOVA) in the Statistical Package for Social Science software (SPSS version 12) and Turkey's Test was used for comparison of means, statistical significance was accepted at $p<0.05$.

RESULTS AND DISCUSSION

Proximate composition of soymilk-tiger nut samples

Table 1 shows the proximate composition of the soy-tiger nut milk. The results obtained showed that there were significant ($p<0.05$) variations in the proximate composition of various samples of soy-tigernut milk samples. It was observed that the ash content ranged from 1.30 to 2.93%, moisture (73.00-81.30%), fibre (0.02-1.83%), fat (2.43-3.93%), protein (5.06-8.48%) and carbohydrate (6.34-16.93%). The moisture was the major component of tigernut-soy milk samples which was about 9.57 to 14.6 times the value of protein and about 4.83 to 11.51 times the value of carbohydrate. Thus, some of the samples contained more protein than carbohydrate. It was also observed that samples with more soy milk had more moisture, even when subjected to more heat treatment. The sample ZAK contained more protein than the other samples.

The ash content varied significantly ($p<0.05$) from 1.30% - 2.93%, with ZAK sample having the highest content of ash, while the sample MAM had the least (1.30%). The ash content of the samples posits the soy-tigernut milk as a mineral-enriched beverage. The ash content of tigernut-soy milk obtained by Orhevba and Bankole (2019) ranged from 0.06-4.38%, implying that the level of substitution of the soy-tigernut milk blends as well as the pasteurization temperature could influence the mineral content of the samples.

It was observed from the surface plot (Figure 4) that increased soy milk addition led to a corresponding decrease in the ash content of the samples, while tiger nut milk inclusion positively influenced the ash content significantly ($p<0.05$). Awonorin and Udeozor (2014) reported that the ash content of soy-tiger nut milk ranged from 0.288 to 0.495%, while Ukwuru *et al.* (2008) obtained the ash content as 1.5%.

The results obtained showed that some of the linear variables (tigernut milk and temperature) positively influenced the ash content of the samples, but the soymilk negatively influenced the ash content significantly ($p<0.05$). Thus, increased soymilk addition led to decreased ash content. It is important to investigate the positive influence of increased temperature of pasteurization on the increase in the ash content of the samples. Orhevba and Bankole (2019) reported a positive effect of ash content due to the increased temperature of pasteurization, although their report suggested that neither soy nor tiger nut milk had effect on the ash content of the samples. However, it must be noted that with their r-squared value of 30.52%, the model could not be substantially explained. The interaction of soy milk with tiger nut milk had a positive effect, while the tiger nut-temperature interaction had a negative effect on the ash content of the soy-tiger nut milk samples.

The quadratic effect of tiger nut milk and soy milk positively influenced the ash content. The r-squared value of 93% showed that the model was fitted.

$$\text{Ash} = 21.02 - 0.705S + 0.1252N + 0.1600T + 0.004504S^2 + 0.000996N^2 + 0.002250SN - 0.00633NT \text{--------------------------} (1)$$

The moisture content of the samples ranged from 73.0 to 81.3%. The sample PAP and ZAK had the least value 73.0%, while sample KAM had the highest value 81.3%. The high moisture content of sample KAM suggests that it had a high nutrient dilution factor. The moisture content of any food is an index of its water activity (Bolarinwa *et al.,*

Table 1. The Proximate composition (%) of soy-tigernut milk samples.

Sample	S:N:T	Ash	Moisture	Fibre	Fat	Protein	Carbohydrate
PAP	58.3:42.7:52	$1.47^{d}\pm0.06$	$73.00^{c}\pm1.00$	$0.07^{d}\pm0.03$	$2.50^{d}\pm0.01$	$5.48^{f}\pm0.01$	$16.93^{a}\pm0.01$
MAM	80:20:54	$1.30^{e}\pm0.10$	$74.00^{c}\pm1.00$	$0.07^{d}\pm0.02$	$2.67^{d}\pm0.06$	$7.63^{b}\pm0.02$	$13.67^{b}\pm0.01$
PAP4	60:40:60	$1.40^{de}\pm0.10$	$76.67^{b}\pm0.0$	$0.02^{e}\pm0.06$	$3.13^{b}\pm0.02$	$6.17^{d}\pm0.02$	$12.38^{c}\pm0.01$
FAD	75:25:45	$1.90^{c}\pm0.10$	$76.33^{b}\pm1.53$	$0.06^{d}\pm0.06$	$3.93^{a}\pm0.01$	$6.83^{c}\pm0.02$	$10.38^{f}\pm0.01$
PAP0	60:40:45	$2.83^{a}\pm0.12$	$76.33^{b}\pm0.06$	$1.50^{b}\pm0.01$	$2.43^{d}\pm0.03$	$5.93^{d}\pm0.02$	$10.98^{e}\pm0.01$
PAP5	75:25:60	$2.50^{b}\pm0.10$	$77.33^{b}\pm0.06$	$1.20^{c}\pm0.01$	$2.87^{c}\pm0.06$	$5.83^{e}\pm0.02$	$10.27^{h}\pm0.01$
KAM	63.9:36.1:52	$2.83^{a}\pm0.06$	$81.33^{a}\pm1.53$	$1.83^{a}\pm0.06$	$2.60^{d}\pm0.01$	$5.06^{g}\pm0.02$	$6.34^{g}\pm0.01$
ZAK	66.7:33.3:45	$2.93^{a}\pm0.06$	$73.00^{c}\pm1.00$	$1.37^{b}\pm0.06$	$2.50^{d}\pm0.02$	$8.48^{a}\pm0.01$	$11.77^{d}\pm0.01$

Values are mean scores ± standard deviation of a triplicate determination. Samples in the same column bearing different superscript differ significantly ($p<0.05$). S = Soymilk (%); N = Tigernut milk (%); T = Pasteurization Temperature (ºC).

2021). This connotes that the KAM sample would be more susceptible to microbial invasion, especially by fungi and mold and thus affect the stability and quality of the samples. With the substantial high moisture content of the samples, the products would require pasteurization and cold storage for shelf-life extension**.**

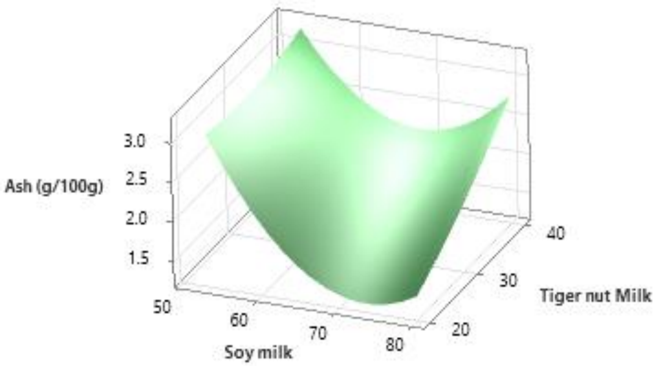

Figure 4. Surface plot of the effect of tiger nut and soy milks on the ash content of the soy-tiger nut milk samples.

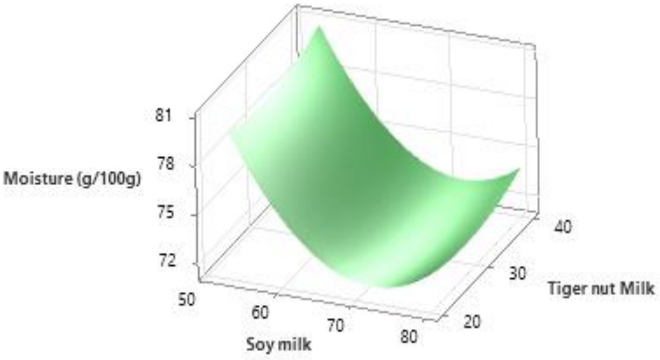

Figure 5. Surface plot of the effect of Tiger nut milk and Soy milk on the moisture content of samples.

$$MC:\ 181.4 - 3.227S - 0.1097N + 0.2667T + 0.02234S^2 + 0.007662N^2 - 0.000000SN - 0.006667NT \text{ ----------------------- } (2)$$

The result of the regression analysis showed that the moisture content was influenced negatively by the addition of soymilk and tiger nut milk, while an increase in pasteurization temperature positively influenced the moisture content of the tiger nut-soy milk samples. Therefore, the inclusion of soy and tiger nut milk would decrease the moisture content of the soy-tiger nut milk, while an increase in temperature would correspondingly lead to an increase in the moisture content of the samples (Figure 5), which agrees with the results obtained by Orhevba and Bankole (2019). Furthermore, the quadratic effect of soy and tiger nut milk positively influenced the moisture content of the samples, while the interaction of soy and tiger nut milk significantly influenced moisture content negatively ($p<0.05$). More so, the interaction effect of tiger nut milk and temperature was negatively influenced ($p<0.05$). The r-squared value of 84% showed that the model was fitted. The moisture content of soy-tiger nut milk obtained by Awonorin and Udeozor (2014) ranged from 81.71 to 86.45%.

The fibre content ranged from 0.02 - 1.83% and differed significantly ($p<0.05$) from each other. The sample PAP4 had the least value of 0.02%, while KAM had the highest value of 1.83%. Most of the values were very low compared to the range of values obtained by Awonorin and Udeozor (2014) which ranged from 0.391 to 0.533%, while some samples PAP0, PAP5, KAM and ZAM had values that were higher than those that have been reported. The presence of high carbohydrates as well as fibre posits the beverage as very important in metabolism and in the development of functional foods (Orhevba *et al.,* 2017).

$$F_iC:\ 11.97 - 0.5898S + 0.4485N + 0.1267T + 0.004391S^2 - 0.002391N^2 - 0.000750SN - 0.005000NT \text{-------------------------} (3)$$

The result of the regression analysis revealed that the fibre content was influenced negatively by the addition of

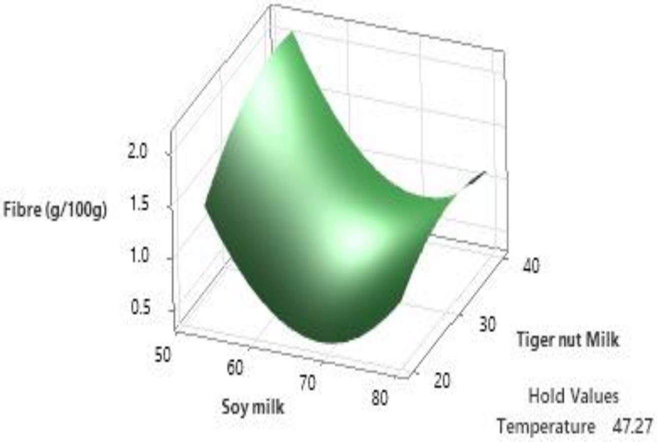

Figure 6. Surface plot of the effect of tigernut and soy milk on the fibre content of samples.

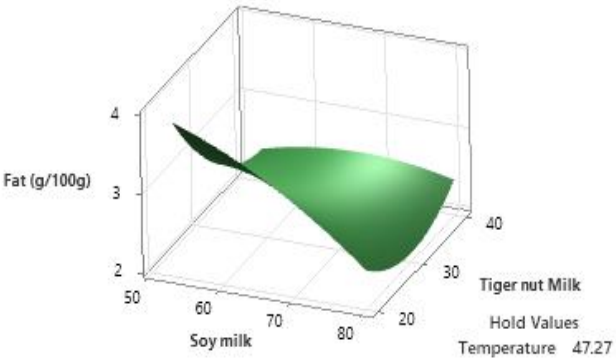

Figure 7**.** Surface plot of the effect of tigernut and soy milk on the fat content of samples.

soymilk, while the other linear variables significantly influenced the fibre content positively. Therefore, the increased addition of soymilk would decrease the fibre content of the soy-tiger nut milk, but the addition of both tiger nuts and an increase in pasteurization temperature would correspondingly lead to the increased fibre content of the samples (Figure 6). Furthermore, the quadratic effect of soy and tiger nut milk positively influenced the fibre content of the samples, while the interaction of soy and tiger nut milk influenced fibre content negatively. Conversely, the interaction effect of tiger nut milk and temperature significantly ($p<0.05$) influenced the fibre content of the samples negatively. The r-squared value of 94% showed that the model was fitted.

Table 1 showed that the fat content of the tiger nut-soy milk samples ranged from 2.43 to 3.93%. There were significant differences ($p<0.05$) in the fat content of the samples which could be attributed to the level of substitution of the samples as well as the effect of pasteurization temperature. Sample PAP0 had the least value of samples 2.43% and sample FAD had the highest value of 3.93%. This could be due to the synergistic effect of soy-tiger nut milk. The results were in support of the work done by Wishh (2006) which stated that the fat content of plain soy milk was about 1.02% and may be varied depending on the ingredients added. Orhevba *et al.* (2017) obtained a range of values of 1.38 - 3.18% for the fat content of tiger nut-soy milk beverages in which an increase in the tiger nut milk led to a corresponding increase in fat content of the samples. Okoroafor and Umeh (2021) observed that the tiger nut-soy milk blends had a fat content value of 2.46 - 3.76%. According to Okoroafor and Umeh (2021), higher fat content in some of their samples was a result of increased tiger nut milk addition.

$$F_aC = 18.61 + 0.007052S - 0.6923N - 0.1733T - 0.000872S^2 + 0.003622N^2 + 0.002750SN + 0.005333NT \text{ -------------------------}(4)$$

However, in this study, all the linear variables negatively influenced the fat content of the samples, while the interaction effect positively influenced the fat contents significantly ($p<0.05$) at a holding temperature of 47.27ºC (Figure 7). The fat content of tiger nut-soy milk blends obtained by Okoroafor and Umeh (2021) was from 3.107 to 4.53%. Other researchers (Roselló-Soto *et al.,* 2019; Duman, 2019; Mohdaly, 2019; Oluwaniti *et al.,* 2009; Belewu and Belewu, 2007) stated that tiger nut oil contained functional ingredients such as phytosterols and tocopherols, including unsaturated fatty acids.

The value of protein in this research ranged from 5.06 - 8.48%. The sample ZAK had the highest value of 8.48%, while the KAM had the least value of 5.06%. Protein is one of the major components of soy-tiger nut milk aside from moisture and carbohydrate. Orhevba and Bankole (2019) reported a protein content that variations in the amount of protein could be due to the effect of heat-processing associated with the pasteurization on the amino acids and obtained a protein content that ranged from 3.28-5.89%. It was believed that tannin could bind protein in a tannin-protein complex to make protein unavailable.

$$P_rC= 43.13 - 0.5317S - 1.205N - 0.1600T + 0.003584S^2 + 0.01192N^2 + 0.004000SN + 0.004667NT\text{--------------------------} (5)$$

The result of the regression analysis revealed that the addition of soy, and tiger nut milks or an increase in temperature decreases the protein content of the samples (See Figure 8 and equation 5). Additionally, the quadratic effect of soy and tiger nut milk positively influenced the protein content of the samples, while the interaction of soymilk and tiger nut milk influenced protein content positively. Also, the interaction effect of tiger nut milk and temperature was positively influenced ($p<0.05$). The r-

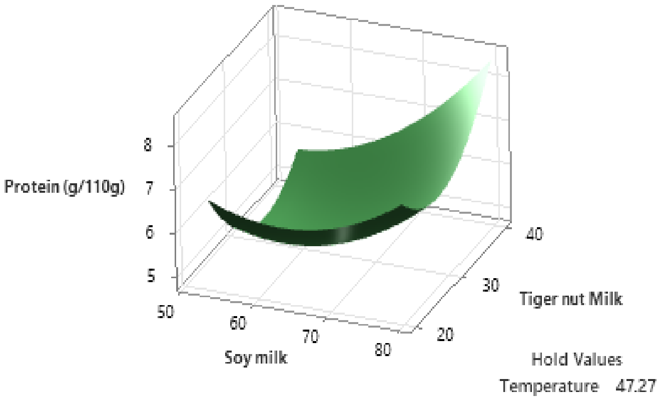

Figure 8. Surface plot of the effect of tigernut and soy milks on the protein content of samples.

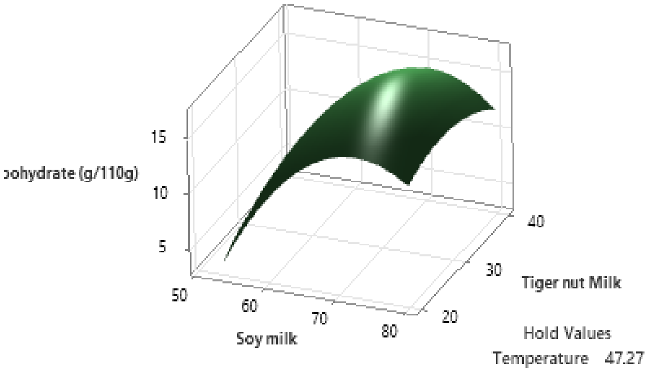

Figure 9. Surface plot of the effect of tigernut and soy milks on the carbohydrate content of samples.

squared value of 47% showed that the model may not be used to explain the effect of the process variables on the protein content of the samples. The variations in the amount of protein could be due to the effect of heat-processing associated with pasteurization on the amino acids. Additionally, tannin could bind protein in a tannin-protein complex thereby making it unavailable. Oluwaniyi *et al.* (2007) posited that total nitrogen reduction leads to a decrease in protein content arising from the destruction of amino acids. An increased content of protein is attributed to tannin-protein complex destruction during processing thereby releasing protein with a consequent increase in the protein content (Abdullahi *et al.,* 2022).

All the samples showed a high content of carbohydrates which ranged from 6.34 - 16.93%. The sample KAM had the least value of 6.34%, while sample PAP had the highest value of 16.93%. There were significant differences (p<0.05) in the carbohydrate content of the samples.

$$CC = 167.3 + 4.680S + 1.232N - 0.1080T - 0.03136S^2 - 0.01759N^2 - 0.06225SN + 0.005033NT \text{-----------------------------} (6)$$

From the regression equation (equation 6) and surface plot (Figure 9), the carbohydrate content was influenced positively by the addition of soy and tiger nut milk, while the other linear variable (temperature) negatively influenced the carbohydrate content of the samples. Therefore, the addition of soy and tiger nut milk would increase the carbohydrate content of the soy-tiger nut milk, while the increased temperature pasteurization led to a decrease in the carbohydrate content of the samples. Furthermore, the quadratic effect of soy and tiger nut milk negatively influenced the carbohydrate content of the samples, while the interaction of soy and tiger nut milk influenced fat content negatively. Conversely, the interaction of tiger nut milk and temperature positively influenced the carbohydrate content significantly (p<0.05). The r-squared value of 80% showed that the model was fitted.

Research by different authors supports the assertion that tiger nut-soy milk blends could serve as a functional food due to the type of carbohydrate found in the beverage (Adebayo-Oyetoro *et al.,* 2017). Adebayo-Oyetoro *et al*. (2017) have shown that tiger nuts can reduce the incidence of diabetics, digestion disorders and lactose intolerance in patients which led Ijarotimi *et al*. (2019) to recommend its use in tiger nut-soy cake blend in the management of diabetes and coeliac diseases.

Mineral composition of soy-tiger nut milk

The mineral composition of various samples of soy-tiger nut milk ranged from 10.20–13.73%, 5.70-7.53%, 2.13-3.53%, 10.27-12.20%, 0.01-0.09%, 10.40-12.80% for calcium, magnesium, iron and phosphorous contents, respectively as shown in Table 2. The results showed that KAM had the least value of 10.20%, while FAD had the highest value of 13.73%. The effect of the different variables on the calcium content of the samples is shown in equation (7).

$$Ca = 2.827 + 0.9751S - 1.234N - 0.2000T - 0.007143S^2 + 0.01414N^2 + 0.000000SN + 0.06667NT \text{---------------------------} (7)$$

The regression analysis showed that the calcium content was influenced positively by the addition of soymilk, while the other linear variables negatively influenced the calcium content of the samples. Therefore, the addition of soymilk would increase the calcium content of the soy-tiger nut milk, but the addition of tiger nut and an increase in temperature would decrease the calcium content of the samples. Furthermore, the quadratic effect of soy milk negatively influenced the soy-tiger nut milk, but the quadratic effect of tiger nut milk positively influenced the calcium content of the samples. However, there was

Table 2. The minerals parameters of soy-tigernut milk samples (%).

Sample	S:N:T	Calcium	Magnesium	Iron	Phosphorous
PAP	58.3:42.7:52	11.83^{e}±0.05	5.70^{b}±0.02	3.13^{b}±0.01	10.83^{d}±0.02
MAM	80:20:54	13.07^{b}±0.02	6.43^{d}±0.01	3.13^{b}±0.05	11.60^{c}±0.01
PAP4	60:40:60	12.80^{c}±0.01	7.03^{bc}±0.01	2.96^{b}±0.04	11.67^{c}±0.02
FAD	75:25:45	13.73^{a}±0.01	6.63^{d}±0.01	2.76^{c}±0.01	12.20^{a}±0.01
PAP0	60:40:45	12.00^{e}±0.01	7.30^{a}±0.01	2.71^{c}±0.01	11.93^{b}±0.05
PAP5	75:25:60	12.50^{d}±0.01	6.93^{c}±0.01	2.52^{d}±0.06	10.87^{d}±0.05
KAM	63.9:36.1:52	10.20^{g}±0.01	5.80^{e}±0.01	2.13^{e}±0.01	10.27^{e}±0.01
ZAK	66.7:33.3:45	11.53^{f}±0.01	7.53^{a}±0.01	3.53^{a}±0.01	11.60^{c}±0.01

Values are mean scores ± standard deviation of a triplicate determination. Samples in the same column bearing different superscript differ significantly ($p<0.05$). S = Soymilk (%); N = Tigernut milk (%); T = Pasteurization Temperature (ºC).

a positive interaction of soy and tiger nut milk on the calcium content of the samples. The r-squared value of 75% showed that the model was fitted.

There are different recommended values of magnesium for each age group in human nutrition. The RDA for children between 1-3 and 4-8 years are 700 mg/day and 1000 mg/day, respectively (NIH, 2021a) for strong bone development in children. Young adults also require calcium for peak bone mass, in addition to bone density maintenance for the reduction in risk of fractures due to ageing. Men and women between the ages of 51-70 and 51-70 years, respectively require a recommended daily intake of 1000 mg/day and 1200 mg/day. Research has shown that calcium intake maintains blood pressure and heart function (Terrar, 2020; Villa-Etchegoyen *et al.,* 2019).

There were significant differences ($p<0.05$) in the magnesium contents of the samples with PAP having the least value of 5.70%, while ZAK had the highest value of 7.53%. The regression equation for magnesium content is as shown;

$$M_gC = 25.83 - 0.2709S - 0.8518N + 0.0600T + 0.001470S^2 + 0.01403N^2 + 0.002000SN - 0.002000NT\text{-------------------------} \quad (8)$$

The regression equation revealed that the magnesium content was influenced negatively by the addition of soymilk and tiger nut milk, while the pasteurization temperature positively influenced the soy-tiger nut milk. Therefore, the addition of soy and tiger nut milk would decrease the magnesium content of the soy-tiger nut milk, but the increased pasteurization temperature decreases the magnesium content of the samples. Furthermore, the quadratic effect of soymilk and tiger nut milk positively influenced the magnesium content of the samples, while the interaction of soymilk and tiger nut milk influenced magnesium content positively. Conversely, the interaction effect of tiger nut milk and temperature was negatively influenced ($p<0.05$). The r-squared value of 56% could not explain the relationship between the magnesium content of the samples and the independent variables. However, it could be asserted that the observed relationship was a chance effect. It could be inferred that the higher composition of soymilk in the present study may have contributed to the increased magnesium content of the samples compared to that obtained by Awonorin and Udeozor (2014) values that ranged from 2.06-3.25 mg/l.

Furthermore, the iron contents of the soy-tiger nut milk samples showed that the KAM had the least value of 2.13%, while ZAK had the highest value of 3.53%, and the samples varied significantly ($p<0.05$) with each other. The regression equation is as shown;

$$Fe = -9.795\% + 0.4219S - 0.09288N - 0.05267T - 0.002931S^2 - 0.000219N^2 - 0.000550SN + 0.001600NT\text{---------------------} \quad (9)$$

It was evident from the regression equation that the iron contents were influenced positively by the addition of soymilk, but the other linear variables negatively influenced the iron content of the samples. This implied that increased soymilk addition would increase the iron content of the soy-tiger nut milk, but the addition of tiger nut milk and increased pasteurization temperature would invariably decrease the iron content of the samples. More so, the quadratic effect of soymilk and tiger nut milk negatively influenced the iron contents of the samples, while the interaction of soymilk and tiger nut milk influenced the iron content positively. The interaction effect of tiger nut milk and pasteurization temperature positively influenced the iron content. The r-squared value of 85% showed that the model was fitted.

Iron is very important for three reasons; the production of haemoglobin, myoglobin and hormone synthesis. The body's need for iron varies in different age groups and is also dependent on whether you are a vegan or not (Villa-Etchegoyen *et al.,* 2019). The body's recommended daily allowance varies from 0.27-27 mg, with lactating mothers needing more iron than others (NIH, 2023; Food Insight, 2022). Ariyo *et al.* (2021) obtained values that ranged from 6.88-9.26 mg/100g for tiger nut milk fortified with date and ginger.

There were significant differences ($p<0.05$) in the phosphorous content of the soy-tiger nut milk samples which ranged from 10.27-12.20%, with KAM having the least value of 10.27%, while the FAD had the highest value of 12.20%. The regression equation is as presented in equation (10).

$$P = 16.28 + 0.4048S - 0.9163N - 0.1733T - 0.003106S^2 + 0.0161N^2 + 0.0000250SN + 0.00433NT \text{---------------------------} (10)$$

The regression equation revealed that the phosphorous contents were influenced positively by the addition of soymilk, while the other linear variables negatively influenced the parameter. Therefore, the addition of soymilk would increase the phosphorous content of the soy-tiger nut milk, but the addition of tiger nut milk and increased pasteurization temperature would lead to a corresponding decrease in the phosphorous content of the samples. The quadratic effect of soymilk and tiger nut milk negatively influenced the phosphorous content of the samples, while the interaction of soymilk and tiger nut milk influenced phosphorous content positively. Conversely, the interaction effect of tiger nut milk and pasteurization temperature positively influenced ($p<0.05$) the phosphorous content of the samples. The r-squared value of 85% showed that the model was fitted. As a very important mineral that is a component of bones, cell membrane structure, teeth, DNA, RNA and as an energy source (adenosine triphosphate, ATP), phosphorous is present in many foods and as a dietary supplement (Heaney, 2012; Calvo and Uribarri, 2018; Henry-Unaeze and Ezeugwu, 2022). Ariyo *et al.* (2021) reported that blends of tiger nut milk, date and ginger had a phosphorous content of 46.54 mg/100g.

Vitamin composition of soy-tiger nut milk

The vitamin composition of soy-tiger nut milk is shown in Table 3. The values of the vitamin contents of the samples ranged from 10.40-12.80 µg, 0.06-1.67 mg/100g, 0.05-1.52 mg/100g, and 0.06-2.30 mg/100g for vitamins A, B_2, C and B_6, respectively. It could be inferred from the regression equation (eqn. 11) that the vitamin A content was influenced positively by the addition of soymilk and tiger nut milk, while increased pasteurization temperature negatively influenced the vitamin A content. The addition of soy and tiger nut milk would consequently lead to increases in the vitamin A content of the samples, but increased pasteurization temperature would decrease the vitamin A content of the soy-tiger nut milk samples.

$$\text{Vit. A} = -32.83 + 1.087S + 0.4580N - 0.03467T - 0.006817S^2 - 0.004283N^2 - 0.003300SN + 0.001067NT \text{-----------------------} (11)$$

The quadratic effect of soy- and tiger nut milk negatively influenced the vitamin A content of the samples, while the interaction of soymilk and tiger nut milk influenced vitamin A contents positively. However, the interaction effect of tiger nut milk and temperature was negatively influenced ($p<0.05$). The r-squared value of 92% showed that the model was fitted. The results obtained in this study align with those of other researchers (Said and Ross, 2014; Ukwuru *et al.,* 2008) whose soy-tiger nut milk formulation ranged from 8.83-30.27 µg. Therefore, the ratio of the blends had effect on the vitamin A contents of the samples. Vitamin A is a fat-soluble vitamin that supports the immune system, eyesight, reproductive health and fetal growth.

There were significant variations in the vitamin B_2 content of the samples. The vitamin B_2 values of 0.05-1.52% were observed in the samples which were within the range of 3.93-2043% obtained by Henry-Unaeze and Ezeugwu (2022). The regression equation is as shown in equation (12);

$$\text{Vit. } B_2 = 17.39 - 01416S - 0.6872N - 0.0123T + 0.000297S^2 + 0.005203N^2 + 0.003500SN + 0.003333NT \text{----------------------} (12)$$

The result reveals that the vitamin B_2 contents of the samples were influenced negatively by the linear variables. Increases in the linear variables would decrease the vitamin B_2 content of the soy-tiger nut milk samples. Both the quadratic and interaction effects of the variables positively influenced the vitamin B_2 content of the samples. Vitamin B_2 (riboflavin) is richly present in natural food sources and works with zinc and niacin to convert vitamin B_6 to its active forms. The r-squared value of 84% showed that the model could be used to explain the relationship between the riboflavin and the independent variables. The RDA values of riboflavin for men and women aged 19 years and above are 1.3 mg/day and 1.1 mg/day, respectively. However, women during pregnancy and lactation require 1.3 mg/day and 1.6 mg/day, respectively. It has been postulated that riboflavin is a key constituent of the coenzymes involved in cell development, energy production, and the breakdown of fats, steroids, and medications in the body. Since they are used immediately after absorption, any excess is readily excreted through urine. The RDA values of riboflavin for men and women aged 19 years and above are 1.3 mg and 1.1 mg/day, respectively (NIH, 2022). However, women during pregnancy and lactation require 1.3 mg/day and 1.6 mg/day, respectively (USDA, 2019). It has been postulated that riboflavin is a key constituent of the coenzymes involved in cell development, energy production, and the breakdown of fats, steroids, and medications in the body. Since they are used immediately after absorption, any excess is readily excreted through urine (NIH, 2022; NIH, 2021b).

The Vitamin C content of the samples differed significantly ($p<0.05$) which could be a result of differences in the ratio of the blends. The result showed that KAM had the least value of 0.05%, while ZAK recorded the highest value 1.52%.

Table 3. The Vitamin contents of soy-tigernut milk samples (mg/100g).

Sample	S:N:T	Vit. A (µg)	Vit. B_2 (mg/100g)	Vit. C (mg/100g)	Vit. B_6 (mg/100g)
PAP	58.3:42.7:52	$12.80^{a}\pm0.01$	$0.08^{e}\pm0.05$	$0.09^{b}\pm0.02$	$2.30^{a}\pm0.01$
MAM	80:20:54	$12.13^{b}\pm0.01$	$0.68^{d}\pm0.01$	$1.03^{b}\pm0.01$	$1.86^{bc}\pm0.05$
PAP4	60:40:60	$11.96^{b}\pm0.04$	$1.60^{a}\pm0.01$	$0.08^{bc}\pm0.01$	$1.95^{b}\pm0.01$
FAD	75:25:45	$11.14^{c}\pm0.06$	$1.23^{b}\pm0.06$	$0.07^{bc}\pm0.01$	$1.83^{bc}\pm0.02$
PAP0	60:40:45	$11.81^{b}\pm0.06$	$1.07^{c}\pm0.06$	$1.07^{b}\pm0.02$	$1.78^{c}\pm0.01$
PAP5	75:25:60	$10.60^{e}\pm0.05$	$0.97^{d}\pm0.01$	$0.09^{bc}\pm0.05$	$0.75^{e}\pm0.02$
KAM	63.9:36.1:52	$10.40^{e}\pm0.02$	$0.06^{f}\pm0.01$	$0.05^{f}\pm0.01$	$0.06^{f}\pm0.01$
ZAK	66.7:33.3:45	$11.80^{b}\pm0.01$	$1.67^{a}\pm0.01$	$1.52^{a}\pm0.01$	$1.34^{d}\pm0.05$

Values are mean scores ± standard deviation of a triplicate determination. Samples in the same column bearing different superscript differ significantly ($p<0.05$). S = Soymilk (%); N = Tigernut milk (%); T = Pasteurization Temperature (°C)

$$\text{Vit. C} = 2.551 + 0.1252S - 0.02536N - 0.04533T - 0.000873S^2 - 0.000652N^2 + 0.000600SN + 0.000967NT \quad (13)$$

The regression equation revealed that the Vitamin C content was influenced positively by the addition of soymilk, while the other linear variables negatively influenced the dependent variable. The inference from the observation was that increased soymilk addition would increase the Vitamin C content of the soy-tiger nut milk, but the increased inclusion of tiger nut milk and pasteurization temperature decreases the ascorbic acid content of the samples. More so, the quadratic effect of soy and tiger nut milk negatively influenced the ascorbic acid content of the samples, while the interaction of soy and tiger nut milk influenced the vitamin C contents positively. The r-squared value of 81% showed that the model was fitted.

The vitamin C (L-ascorbic acid) as a water-soluble vitamin, is naturally present in some foods and as a dietary supplement that cannot be endogenously synthesized by humans (Sfayhi-Terras *et al.,* 2021). It is required for the biological synthesis of collagen (which plays a vital role in wound healing), L-carnitine, and certain neurotransmitters, as well as in protein metabolism. It is important to note that vitamin C plays a vital role as a physiological antioxidant that has the ability to regenerate other antioxidants within the body, such as α-tocopherol (Osungbade *et al.,* 2016).

There were significant differences ($p<0.05$) in the vitamin B_6 content of the samples which could be attributed to the differences in the blend ratio of the tiger nut and soy milk, as well as the effect of pasteurization temperature (egn. 14). The result revealed that KAM had the least value of 0.06 mg/100g of vitamin B_6, while PAP had the highest value of 2.30 mg/100g. The samples MAM and FAD showed no significant differences ($p>0.05$).

$$\text{Vit. } B_6 = -21.08 + 0.9280S - 0.209N - 0.1980T - 0.006568S^2 - 0.000357N^2 - 0.000675SN + 0.005667NT\text{-----------------------} (14)$$

It was observed from the regression equation that the vitamin B_6 content of the samples could be increased by increased addition of soymilk, while tiger nut milk addition and increased pasteurization temperature would decrease the vitamin B_6 content of the samples (eqn. 14). Additionally, the quadratic effect of soymilk and tiger nut milk negatively influenced the Vitamin B_6 content of the samples, just as the interaction of soymilk and tiger nut milk influenced vitamin B_6 content negatively. Conversely, the interaction effect of tiger nut milk and temperature positively influenced the vitamin B_6 content ($p<0.05$). The r-squared value of 81% showed that the model was fitted.

According to Bird (2018), vitamin B_6 is a complex molecule which is essential for the maintenance of human health as it plays a vital role in anti-inflammatory-related chronic diseases such as cancer. The role of vitamin B_6 in influencing the post-translational modification of important proteins responsible for inflammatory reactions, metabolism and immune system modulation has been established.

Functional properties of soy-tiger nut milk samples

The functional properties of soy-tiger nut milk were evaluated with significant variations ($p<0.05$) observed in both the water absorption capacity and solubility index (Table 4). The water absorption capacity of the samples ranged from 51 to 69.67 mg/100g, while the value of the solubility index ranged from 15.23 to 16.67. The sample KAM had the least water absorption capacity, while the sample PAP had the highest value.

$$\text{WAC} = -188.4 + 7.249S + 1.229N - 1.133T - 0.04785S^2 - 0.004465T^2 - 0.007500SN + 0.04000NT\text{--------------------------} (15)$$

Heat-moisture treatment has been reported to cause increases in the water absorption capacity of foods (Sfayhi-Terras *et al.,* 2021). The regression equation revealed that the water absorption capacity was influenced positively by the other linear variables, except the pasteurization temperature which had a negative influence

Table 4. Functional parameters of soy-tigernut nut.

Sample	S:N:T	Solubility index (mg/100g)	Water absorption capacity (mg/100g)
PAP	58.3:42.7:52	16.67^{a}±1.53	69.67^{a}±1.52
MAM	80:20:54	16.67^{a}±0.02	64.33^{b}±1.15
PAP4	60:40:60	16.30^{a}±0.01	62.33^{c}±0.58
FAD	75:25:45	16.10^{b}±0.01	56.00^{e}±1.00
PAP0	60:40:45	16.07^{ab}±0.02	54.00^{f}±1.00
PAP5	75:25:60	16.33^{a}±0.06	53.67±1.53^{f}
KAM	63.9:36.1:52	15.23^{b}±0.02	51.00^{g}±1.00
ZAK	66.7:33.3:45	15.60^{ab}±0.01	60.33^{d}±0.06^{d}

Values are mean scores ± standard deviation of a triplicate determination. Samples in the same column bearing different superscript differ significantly ($p<0.05$). S = Soymilk (%); N = Tigernut milk (%); T = Pasteurization Temperature (ºC).

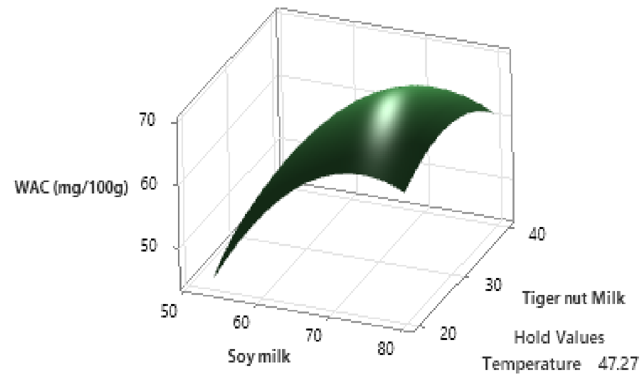

Figure 10. Surface plot of the effect of tigernut and soy milks on the water absorption capacity of samples.

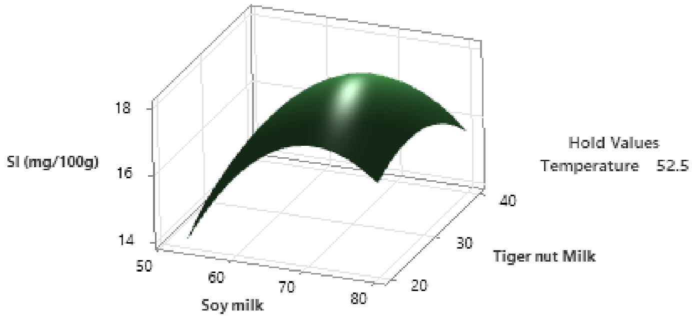

Figure 11. Surface plot of the effect of tigernut and soy milks on the solubility index of samples.

on the samples. Consequently, the addition of soy and tiger nut milk would independently increase the water absorption capacity of the soy-tiger nut milk, while increased pasteurization temperature would decrease the water absorption capacity of the samples (Figure 10). Furthermore, the quadratic effect of soymilk and tiger nut milk negatively influenced the water absorption capacity of the samples, while the interaction of soymilk and tiger nut milk influenced water absorption capacity negatively (Figure 7).

The solubility index of the soy-tiger nut milk exhibited significant variations ($p<0.05$), with the sample KAM having the least solubility index, while PAP had the highest value. PAP4 and PAP5 showed significant differences ($p<0.05$). The solubility index measures the volume of undissolved components (sediments) that are left after healthy methodical remoistening or rehydration.

$$SI = -45.83+1.534S + 0.6275N + 0.02000T - 0.01046S^2 - 0.008292N^2 - 0.002250SN + 0.000000NT \text{------------------------} (16)$$

The solubility index was significantly ($p<0.05$) influenced positively by the linear variables (R^2-value of 85%) (Figure 11). The pasteurization temperature of the soy-tiger nut milk significantly led to a high solubility index due to the fact that solids exhibit solubility at temperatures above 20ºC as the average kinetic energies of solvent molecules increase and dislodge more particles from the surface of solutes.

The high solubility index shown by both the tiger nut and soy milk could be due to the high soluble sugars in the samples. The plot of the interaction between the linear variables showed that the optimum solubility index could be fitted at various levels (Figure 12), especially at a blending ratio of 70% soy milk and 30% tiger nut milk formulation.

The quadratic effect of soy and tiger nut milk negatively influenced the solubility index of the samples, while the interaction of soy and tiger nut milk influenced the solubility index negatively. However, the interaction effect of tiger nut milk and temperature was positively influenced ($p<0.05$) significantly. The r-squared value of 87% showed that the model could explain the process.

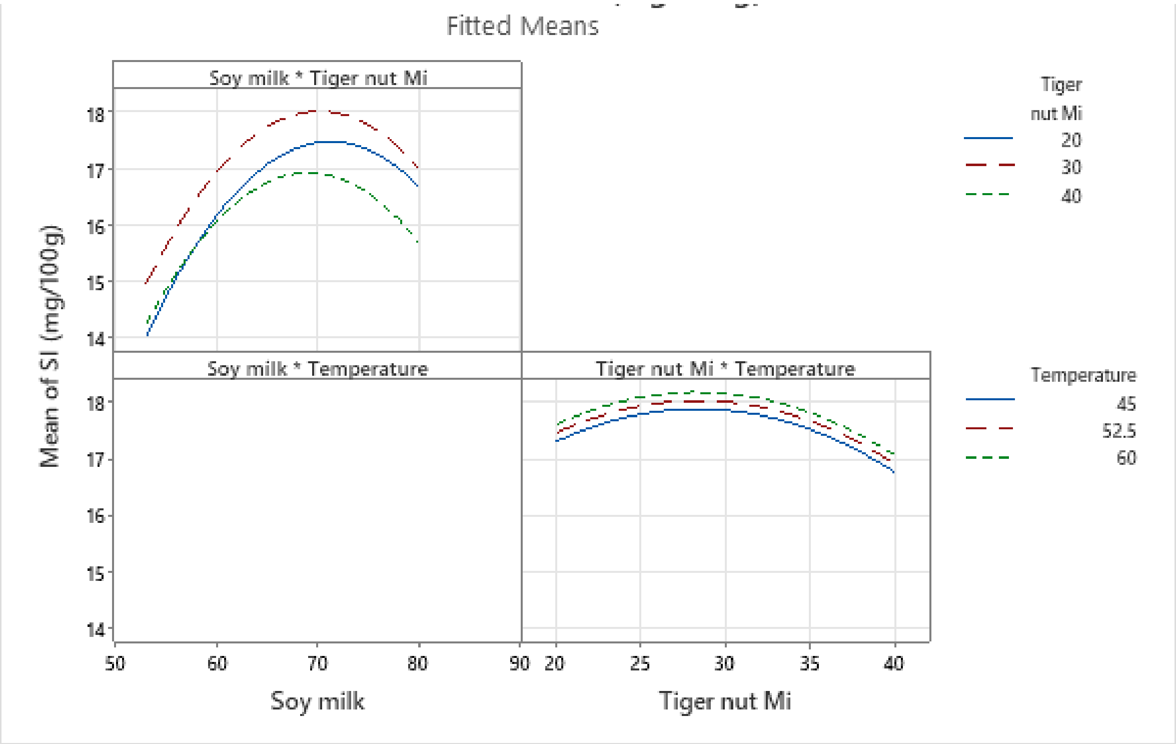

Figure 12. Interaction plot of the effect of tigernut and soy milks on the solubility index of samples.

Table 5. The phytochemical parameters.

Sample	S:N:T	Flavonoids (mg/100ml)	Phenols (mg/100ml)
PAP	58.3:42.7:52	6.23^{e}±0.01	11.60^{f}±0.01
MAM	80:20:54	7.37^{c}±0.02	12.43^{d}±0.02
PAP4	60:40:60	6.87^{d}±0.02	12.70^{bc}±0.02
FAD	75:25:45	7.40^{c}±0.01	12.03^{c}±0.02
PAP0	60:40:45	8.50^{a}±0.01	12.93^{c}±0.05
PAP5	75:25:60	8.10^{b}±0.02	12.47^{cd}±0.02
KAM	63.9:36.1:52	6.90^{d}±0.01	11.87^{e}±0.05
ZAK	66.7:33.3:45	8.03^{b}±0.02	12.96^{a}±0.05

Values are mean scores ± standard deviation of a triplicate determination. Samples in the same column bearing different superscript differ significantly ($p<0.05$). S = Soymilk (%); N = Tigernut milk (%); T = Pasteurization Temperature (ºC).

Phytochemical composition of soy-tiger nut milk samples

The phytochemical composition of soy-tiger nut milk ranges from 6.23 - 8.50 mg/100g and 11.60 - 12.96 mg/100g of flavonoids and phenols, respectively, as shown in Table 5. The result showed that PAP had the least flavonoid content of 6.23 and PAP0 had the highest value of 8.50. However, other samples differed significantly ($p<0.05$).

$FlC = 15.26 - 0.2087S - 0.2806N + 0.1733T + 0.001491S^2 + 0.01176N^2 - 0.000750SN - 0.007333NT$------------------------- (17)

The result of the regression reveals that the flavonoids were influenced negatively by the addition of soymilk, while the other linear variables were negatively influenced. Therefore, the addition of soymilk would decrease the flavonoid of the soy-tiger nut milk, while the addition of both tiger nut and temperature increase led to a corresponding increase in the flavonoid of the samples (Figure 13).

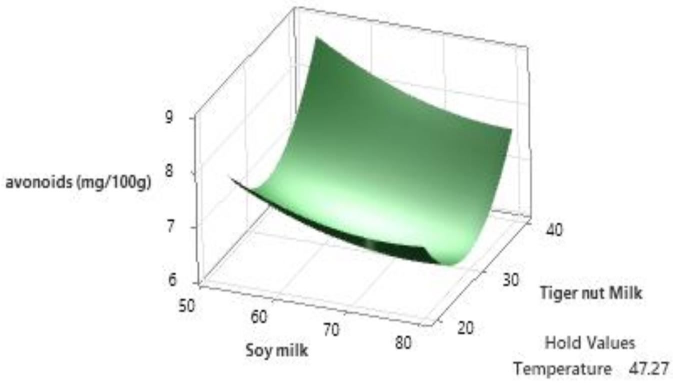

Figure 13. Surface plot of the effect of tiger nut and soy milks on the flavonoids content of samples.

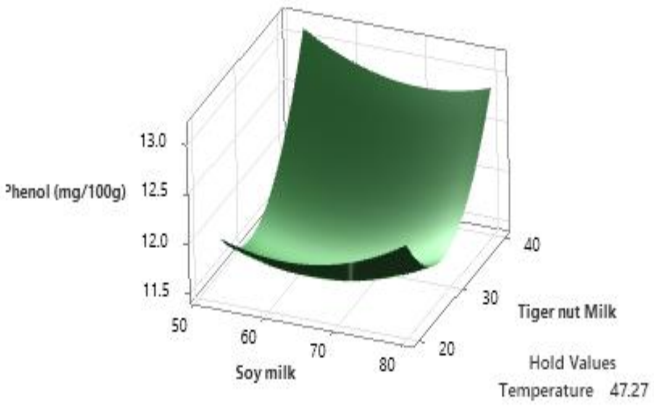

Figure 14. Surface plot of the effect of tigernut and soy milks on the phenol content of samples.

Furthermore, the quadratic effect of soymilk and tiger nut milk positively influenced the flavonoids of the samples, while the interaction of soymilk and tiger nut milk influenced flavonoids negatively. Conversely, the interaction effect of tiger nut milk and temperature was negatively influenced ($p>0.05$). The r-squared value of 98% showed that the model was fitted.

The result showed that PAP had the least value of 11.60 mg/100g of phenol and ZAK had the highest value of 12.96 mg/100g of phenol, while significant differences ($p<0.05$) were observed in other samples such as MAM and FAD.

$$\text{Phenol} = 19.21 - 0.1422S - 0.3219N + 0.04667T + 0.001302S^2 + 0.008198N^2 - 0.001000SN - 0.00133NT \text{ ----------------------- } (18)$$

From the regression analysis, the result reveals that the phenol was influenced negatively by the addition of soymilk while the other linear variables were negatively influenced. Therefore, the addition of soymilk would decrease the phenol of the soy-tiger nut milk, while the addition of both tiger nut and temperature decrease led to the corresponding decrease in the phenol of the samples (Figure 14). Furthermore, the quadratic effect of soymilk and tiger nut milk positively influenced the phenol of the samples, while the interaction of soymilk and tiger nut milk influenced phenol negatively. Conversely, the interaction effect of tiger nut milk and temperature was negatively influenced ($p<0.05$). The r-squared value of 100% showed that the model fitted.

Sensory evaluation of soy-tiger nut milk

The results for sensory evaluation of soy-tiger nut milk are shown in Table 6 which showed significant variations ($p<0.05$) in the panel's evaluation of the sensory attributes. Sensory attributes affect the perception of food to be consumed and play a major role in determining consumer acceptance of food products. The aroma, appearance, colour, texture and overall acceptability of the samples ranged from 4.65 - 7.65%, 4.35 - 7.35%, 3.95 - 7.55%, 3.90 - 7.40%, 4.95 - 7.75%, respectively.

The result showed that sample PAP5 (4.65) had the least value for Aroma, while sample FAD (7.65) had the highest value. The sample with 60% soymilk, 40% tiger nut milk and pasteurized at 45ºC had the best aroma (like very much), compared to the other samples, while PAP5 formulated by blending 75% of soymilk with 25% of tiger nut milk and pasteurized at 60ºC was rated 4.65 (dislike slightly), as evaluated by the panellist. Thus, high pasteurization temperature may have impacted a burnt taste to the product which may have negatively influenced the panellists.

The appearance of the samples significantly differed ($p<0.05$) (Table 6). The samples PAP0, PAP5 and KAM were rated 4 (disliked slightly) by the panellist and thus had no significant differences ($p>0.05$) between them, while significant differences ($p<0.05$) were observed between them and other samples. Moreover, samples MAM and ZAK were ranked 7 (liked very much) by the panellists based on their appearance. From the result obtained as shown in Table 6, sample PAP0 had the least value for appearance (4.35%). These could be due absence of stabilizers in the samples, blend ratio or the effect of pasteurization temperature. It was observed that samples with stabilizers were ranked better by the panellists.

In terms of colour rating, the panellists ranked PAP5 the least (dislike moderately), followed by KAM and PAP0 (dislike slightly), while samples MAM and FAD were ranked highest (liked moderately). The variations of the colour rating were significant ($p<0.05$). Almost the same trend was observed in the rating of the samples according to their texture, while the overall acceptability rating showed that MAM, FAD and ZAK were moderately liked by the panellists. The sample PAP0 ranked least in the general acceptability rating. Therefore, soy-tiger nut

Table 6. Sensory properties of soy-tigernut milk samples.

Sample	S:N:T	Aroma	Appearance	Colour	Texture	Overall acceptability
PAP	58.3:42.7:52	6.00^{bc}±2.71	5.95^{b}±2.14	6.10^{bc}±2.55	6.15^{ab}±2.16	6.25^{bc}±2.31
MAM	80:20:54	6.80^{ab}±2.50	7.20^{a}±2.21	7.00^{ab}±2.29	7.40^{a}±1.57	7.75^{a}±1.59
PAP4	60:40:60	4.80^{de}±2.24	5.55^{bc}±2.11	5.50^{cd}±2.52	5.35^{ac}±2.06	5.30^{cd}±2.20
FAD	75:25:45	7.65^{a}±1.14	7.35^{a}±1.27	7.55^{a}±1.47	7.20^{a}±1.88	7.55^{a}±1.09
PAP0	60:40:45	5.50^{c}±1.88	4.35^{c}±1.73	4.05^{e}±1.85	5.35^{bc}±2.37	4.95^{c}±2.06
PAP5	75:25:60	4.65^{d}±1.89	4.55^{bc}±1.76	3.95^{f}±1.50	3.90^{c}±1.83	5.20^{cd}±1.58
KAM	63.9:36.1:52	5.00^{de}±2.10	4.55^{bc}±1.67	4.05^{ef}±1.79	4.95^{bc}±2.16	5.25^{cd}±1.52
ZAK	66.7:33.3:45	7.20^{a}±2.46	7.25^{a}±2.45	6.65^{b}±2.46	7.00^{ab}±2.24	7.25^{ab}±1.80

Values are mean scores ± standard deviation of a triplicate determination. Samples in the same column bearing different superscript differ significantly ($p<0.05$). S = Soymilk (%); N = Tigernut milk (%); T = Pasteurization Temperature (°C).

Table 7. Cultural and morphological characteristics of bacterial isolate.

Isolate code	Size	Opacity	Edge	Surface	Elevation
58.3:42.7:52	2 mm	opaque	Entire	Mucoid	Raised
80:20:54	3 mm	opaque	Entire	Mucoid	Raised
60:40:60	2 mm	opaque	Entire	Mucoid	Raised
75:25:45	3 mm	opaque	Entire	Mucoid	Raised
60:40:45	3 mm	opaque	Entire	Mucoid	Raised
75:25:60	1 mm	Opaque	Entire	Mucoid	Raised
63.9:36.1:52	1 mm	Opaque	Entire	Mucoid	Raised
66.7:33.3:45	1 mm	Opaque	Entire	Mucoid	Raised

milk samples could be produced that would be moderately liked by the consumers.

Microbial characterization

The results of the microbial analysis showed that the microbes isolated from the different samples had the same morphological characterizations, although their size ranges differed (Table 7). The isolates were opaque, entire, mucoidal and raised with a size range of 1 - 3 mm. Furthermore, samples with more soy to tiger nut milk ratio subjected to minimal heat treatment were observed to have more isolate size. Therefore, pasteurization temperature had effect in reducing the size of the isolated microorganisms, while more soymilk addition relative to tiger nut milk increased the size of microbial isolates.

The results of the biochemical tests showed that the microbial isolates of the sample PAP (1) are rod-shaped, and gram-negative. The reactions of the isolates to citrate, oxidase, lactase, gas, indole and coagulase were negative but reacted positively to glycosidase, lactase, hydrogen sulphide, Voges-Proskauer, catalase and mortality reaction with a proble suspicion of the presence of Bacillus species in the sample. The triple sugar test was classified as A, while the test for agar butt was classified as B.

The microbial isolate suspected to have been isolated in the sample MAM were micrococcus species as they were found to be cocus-shaped and reacted negatively to the following reactions; citrate, glucose, hydrogen sulphide, indole, coagulase and mortality tests. Other microorganisms suspected to have been isolated include *Klebsiella spp, Stapphylococcus spp., Corynebacterium spp., Bacillus spp., Stappylococcus and Pseudomonas* for samples PAP4, FAD, PAP0, PAP5, KAM and ZAK, respectively (Table 8).

Table 9 shows the colony characterization and morphological identification of fungi isolates in the soy-tiger nut milk samples. It was evident that that most of the fungi species identified were *Aspergillus species* which were identified in samples KAM (7) and ZAK (8). There were also *Saccharomyces, Rhizopus, Fusarium, Penicillium* and *Candida species.* The presence of the toxic organisms would have been caused by the processing methods employed, the type of equipment used or the inadequate pasteurization temperature. Onovo and Ogaraku (2007) have identified *Bacillus subtilis, Stapphylococcus aureus, Aspergilus flavus., A. niger, Fusarium solani, Saccharomyces cerevisiae, S. fubiligera* and *Candida pseudotropicalsi* in tiger nut milk.

Table 8. Biochemical characteristics of bacterial isolates.

isolates	shape	GR	Citrate	Glucose	Oxidase	Lactose	TSI	AB	H_2S	Gas	MR	VP	indole	Catalase	mortality	Coagulase	Probale organisms
1	rod	-	-	+	-	+	A	B	+	-	-	+	-	+	+	-	Bacillus spp
2	coci	+	-	-	+	+	A	B	-	-	+	-	-	+	-	-	Micrococcus spp
3	coci	-	+	+	+	+	A	B						+	-	-	Klebsiella
4	coci	+	+	-	-	+	B	A	-	-	+	-	+	+	+	-	Staphylococcus spp
5	coci	-	+	+	-	-	A	A	-	-	+	-	-	+	-	-	Corynebacterium
6	rod	-	-	+	+	+	A	B	-	+	-	-	+	+	+	-	Bacillus sp
7	rod	+	+	-	-	+	B	B	+	-	-	+	+	+	-	-	Staphylococcus spp
8	rod	-	-	+	-	+	A	A	+	-	+	+	-	+	-	-	Pseudomonas spp

MR = methyl red; VP= voges proskaller; TSI = Tripple sugar iron; A = Acid; B = base; H_2S = Hydrogen sulphide; AB = Agar butt; GR = Gram reaction.

Table 9. Colonial characterization and morphological identification of fungal isolates.

isolate code	Colonial features	Morphology	Probable organism
1	Yellowish and powdery colonies	Circular spore, no hyphae and conidia	*Candida* sp
2	Pink and cottony colonies	*Microconida* are ovoid in shape. *Microconidia* are borne on phialides on branched *conidiosphores*, septate fusiform, slightly curved and pointed at both ends is present	*Fusarium* sp
3	Green and velvet	Colonies are smooth and ellipsoidial. *Conidiosphores* are smooth and short. Mycelia are arranged irregularly with branches of various lengths	*Penicillium* sp
4	Flat, creamy and mucoidy colonies	Branched *conidiosphores* with smooth conidia but with a rough wall	*Fusarium* sp
5	Whitish colonies growing rapidly and filling the petri dish with dense cottony mycelium and becoming brownish-black with ag	Non-septate mycelia. *Sporangiosphores* are smooth walled. *Sporangia* and *columella* are subglobose. *Sporangiospores* are ovoid in shape	*Rhizopus* sp
6	Dense white budding colony separated by a brown line	Non septate hyphae with large sporangia heads having numerous *sporangiospores*	*Sacccharomyces* sp
7	Bluish green colonies with a suede like surface	Conidia heads are large, globose, dark-brown and biseriate, conidia are globose and rough walled. *Conidiosphores* are smooth walled	*Aspergillus* sp
8	Black colonies with white edges	Conidia heads are large, globose, dark-brown and biseriate, conidia are globose and rough walled. *Conidiosphores* are smooth walled	*Aspergillus* sp

Conclusion

The study has shown the production and quality evaluation of soy-tiger nut faux milk blends which are rich in soluble carbohydrates, proteins, fats, and ash but low in fibre (0.02-1.8%). In mineral composition, calcium and phosphorous content showed high values which ranged from 10.20 – 13.07 mg/100g and 10.27 – 12.20 mg/100g, respectively, with low iron content that ranged from 2.13 to 3.53mg/100g.

The vitamin content of the soy-tiger nut milk samples was low comparable to the recommended daily allowance. The result of phytochemicals showed that the samples are rich in flavonoids and phenols. There were significant differences (p<0.05) in the functional properties of the samples, with water absorption capacity and solubility index having values that ranged from 51.00 to 67.67 mg/100g and 15.23 to 16.67 mg/100g.

The result for sensory evaluation showed that significant differences (p<0.05) were observed. It may be concluded that samples ZAK, FAD and MAM showed moderate acceptance by the panellists, which posits it as a very good beverage for consumers.

In microbial load, the bacteria organism isolated were *Bacillus spp, Microccoccus spp, Klebsiella spp, Staphylococcus spp, Corynebacterium spp* and *Pseudomonas spp,* while the fungi isolates include *Candida spp, Fusarium spp, Penicillium and Rhizopus spp* and *Saccharomyces*.

However, the large microbial isolates in the beverage may posits to suggest that appropriate pasteurization temperature could be determined to ensure the elimination of most of the pathogenic organisms isolated from the beverage. However, it had been noted that temperatures above 50ºC may be detrimental to the sensory, as well as nutritional composition of the soy-tiger nut beverage.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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