

Natural tropical and chemical preservatives effects on shelf-life and hedonic acceptability of tiger-nut milk and soymilk under storage conditions: A review

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Abstract

Soy milk and tiger-nut milk were processed into natural fresh beverage. The problem of preservation of these milks as vegetable milks samples is common in many countries. In such situations, many research were conducted. This article proposes a review on quality of some different preservatives used to preserve and extend shelf-life of tiger-nut milk and soymilk under storage conditions. The effects of preservatives on the microbiological quality and the acceptability of milks were investigated. It arises that tiger-nut milk and soymilk undergoes various changes during storage conditions. In spite of the dangerousness of chemical preservatives for human health, the most preservatives used were the chemical or synthetic one. Unfortunately, after a long period of storage of milks in that it were added, the hedonic acceptability is virtually unacceptable. The shelf-life of soymilk or tiger-nut milk increased by different preservatives if stored at refrigerated condition. These information will be guiding the globally of nutritionist and manufacturing unit the appropriate preservatives for extending and preserve tiger-nut milk and soymilk.

Keywords: tiger-nut milk, soymilk, preservatives, food safety, shelf-life, acceptability

Introduction

Milk has been recognized as an important food for infant and growing children ^[1] because milk is an excellent source of nutrients such as vitamins, amino acids, fats, minerals, proteins and sugar, making it an excellent medium for microbial proliferation ^[2]. In developing countries, the cost of dairy milk and their products are beyond the reach of common man. The high cost of milk in developing countries has led to the development of alternative source of milk from plant minerals. An inexpensive substitute in the form of a milk or beverage made from locally available plant foods, high in protein, with satisfactory quality milk could play an important role to reduce protein malnutrition ^[3]. In view of the scarce milk supply in various countries and the ever increasing gap between the requirement and population, efforts have been made over the years to develop alternative milk-like products from vegetable sources ^[4]. Though undervalued in the past, milk from plant sources are key ingredient in the diet of African countries. Recently, researchers have shown strong interest in these milk sources due to their high nutritional values and economic potentials. It is worthy repeating that milk sources from plants are seen as a radiating hope as well as an ally in the fight against hidden hunger. Among milks from plant, seeds, soymilk and tiger-nut milk have been received great interest and high attention in the investigations on milk substitutes.

Soy milk (*Glycine max*) is a healthy drink and is important for people who are allergic to cow milk protein and lactose ^[5]. Tiger-nut milk from *Cyperus esculentus* is mainly consumed in several countries of Africa and attract all the categories of people ^[6] ^[7]. The two milks are recommended for it beneficial effect related to protein content, the quality of the fatty acid, the content of vitamins, minerals and their capacity to prevent certain diseases ^[8] ^[9]. Also, delicious, plants milks are well appreciated by consumers and is consumed like regular drink and replacement milk for dairy milk ^[10]. It is more consumed in hot periods of the year ^[6] ^[11].

As a local beverage, produced by traditional methods, offer only a short lifespan. The milks are underutilized due to its short shelf-life ^[12]. The short shelf-life of raw tiger-nut milk hinders widespread consumption of the beverage due to the deteriorating effects of some microorganisms on the milk ^[13]. Bacterial contamination of milk analogue can originate from different sources: bad water, air, preparation equipment, unhygienic of the handler, poor post-pasteurization handling such as bottling and storage systems, among others. Pasteurization is effective in eliminating all but the thermotolerant microorganisms of the genera *Microbacterium*, *Micrococcus*, *Streptococcus*, *Lactobacillus*, *Bacillus*, *Clostridium*, the coliform, and occasionally some Gram-negative rods ^[14]. Psychrotrophs can grow at refrigeration temperatures below 7°C, produce

enzymes, toxins and other metabolites [14] and contribute to high standard plate counts in both raw and pasteurized milk analogue. Unlike most dairy products, milk analogue is currently handled and stored at low temperatures; these organisms hinder efforts to increase the shelf-life of pasteurized product [15]. Pasteurization cannot guarantee the absence of microorganisms, when they are present in large numbers in raw material or due to post-pasteurization contamination. Examination for the presence and number of specific micro-organisms is, therefore, an integral part of any quality control or quality assurance plan.

One of the main concerns in the food process industry is also the survival and growth of pathogens, causing bacterial spoilage and human infections, and affecting the health and safety of consumers [16]. There were many attempts to industrialize the locally prepared tiger-nut milk and soymilk, but the inability to preserve the milks for a long time without spoilage has been a major problem. Food products are an excellent environment for growth of pathogenic microorganisms, which may cause food-borne diseases. Quality and shelf-life of food products depend greatly on the properties of microorganisms contaminating the food [17]. Bio-deterioration of the milk is influenced by factors like temperature and microbial load. A large number of microorganisms such as coliforms, mesophilic aerobic bacteria, yeasts and moulds are responsible for the spoilage and it can produce undesirable changes in the milk. These micro-organisms are the limiting factors of the keeping quality of milk. To optimizing storage conditions, the rotting process can be postponed by adding preservatives or applying modern techniques. For that, processing methods and preservatives were developed to maintain the color, value and food acceptability [18]. Consumers have a growing

preference for natural and fresh tasting [19], convenient, healthy, palatable, microbiologically safe [20], high quality, free from chemical preservatives and additives foods [19] [20] with a clean label and an extended shelf-life has increased [19]. The preservatives in food preparations inhibit or retard the growth of microorganisms which reduces the deteriorative effects of microorganism on food [21]. However, this present work was performed to investigate the efficiency of various natural and chemical preservatives extending shelf-life of tiger-nut milk, soymilk during storage and its acceptability consumer's.

Methodology

A literature review was conducted on chemical and natural preservatives used in soymilk and tiger-nut milk associated to processing treatment to preserve the quality of the milks storage at ambient temperature and under refrigeration and it acceptability. The search was carried out in Google scholar databases, Crossref Metadata Search, Agora using the following keyword such us: tiger-nut milk, soymilk, preservatives, food safety, shelf-life and acceptability. The review made over the period from October 2018 to Mai 2019.

Literature Review

1.1 Preservatives allowed in milks and milks products for conservation

Many food products are perishable by nature and require protection from spoilage during their preparation, storage, and distribution to give them the desired shelf-life. Some food additive standardized such us conservator, antioxidant set by Codex alimentarius are presented (table 1).

Table 1: Some preservatives and limit dose in milks products [22]

Preservatives	Function	Codex Stan 192-1995 [Dose limit (mg/kg)]
Sorbic acid (E200)	Conservator	300 – 1000
Potassium sorbate (E202)	Conservator	3000
Calcium sorbate (E203)	Conservator	3000
Nisin (E234)	Conservator	12.5
Ascorbic acid (E300)	Antioxidant	0.5
Alginic acid	Stabilizing	BPF
Citric acid	Antioxidant	BPF
Benzoic acid	Conservator	300
Sodium-benzoate	Conservator	300
Potassium-benzoate	Conservator	300
Calcium-benzoate	Conservator	300
Dioxyd carbonate	Conservator	BPF
Ethyl hydroxybenzoate	Conservator	120
Methyl hydroxybenzoate	Conservator	120

1.2 Preservatives used in soymilk and tiger-nut milk for conservation tests

The research reported the use of various preservatives such as chemical preservatives and natural tropical to overcome the problem of soymilk or tiger-nut milk biodeterioration. For that, chemical preservatives such as azidiol or sodium azide/chloramphenol [23], sodium azide [24], sodium-benzoate, sodium metabisulphite only or combined [25] were explored. Other studies reported potassium-meta-bi-sulphite (KMS), sodium-benzoate and potassium sorbate [26]. As natural preservatives ginger (*Zingiber officinale*) and garlic [27], citric acid, garlic, ginger, and clove were test with little success [28]. The use of cloves extract (*Syzygium*

aromaticum) and guinea-pepper (*Xylopi aethiopica*) to preserve soymilk was reported [21]. Some research reported also the addition of nisin and nipagin complex esters in soymilk [29] and *Aframomum danielli* spice powder to every 200 mL of the blend [30]. Making vegetable milks available throughout the year because of their short shelf-life, man has developed the combination of methods including germinating, soaking, fermentation, blanching, sterilization, dehydration, ultrasound or ultrasonic to prolong the storage life of products to preserve them. Therefore, some combined processing has develop with thermal treatment such as pulsed electric field [31] [32] [33] and high pressure processing [34]. Recently researchers are proposing the use of milder

heat treatments in combination with other hurdles such as natural antimicrobials, preferably of natural origin [35] [36].

1.3 Microbiological quality of soymilk, tiger-nut milk stored at different conditions

The growth of microorganisms may be attributed to the cells of lactic acid bacteria that might have survived through processing treatments. The fungal and viable count of tiger-nut milk samples calls for concern. This has been linked to the spoilage of the storage microflora (table 2, 3, 4, 5). The microbiological quality content of the tiger-nut milk pasteurized and treated with sodium azide was low at the twelve day period of storage in total viable count (31±12 cfu/mL) and 21±15 cfu/mL for fungi count [24]. There is more pronounced reduction in total microbial count in

soymilk samples treated with clove extract (1.92x10⁵ cfu/mL) as compare to the samples treated with guinea pepper extract (2.70x10⁵ cfu/mL) reported by [21]. Bacterial load of pasteurized tiger-nut extract stored at 4±2°C of most treatments were fairly good up to day 8 except garlic, citric acid and garlic and ginger formulated that had extended fairly good beyond day 8. This met with International Standard Organization (ISO) [37] statement that milk after storage at 5°C its count at 21°C should be less than 10⁵ cfu/mL [38] states that milk sample containing 5.00x10³ cfu/mL of bacteria is classified as good for consumption, 1.00x10⁴ to 4.00x10⁵ cfu/mL is fairly good, 2.00x10⁶ is passable, while over 2.00x10⁷ cfu/mL is bad for consumption.

Table 2: Total viable bacterial count of different soymilk samples [39]

Samples	Preservatives			Total viable count (log cfu/g)
	Potassium meta-bi sulphite (KMS)	Sodium- benzoate	Potassium Sorbate	
1	0	0	0	5.294
2	300	0	0	5.293
3	0	350	0	5.248
4	0	0	350	5.277
5	250	250	200	5.120

Table 3: Microbial load of tiger-nut milk stored at ambient temperature [24]

Treatments	Period of storage (day)	Microbial count (cfu/mL)	
		Total viable count	Fungal count
Pasteurized + 0.08% sodium benzoate	1	1.33x10 ⁴	1.93x10 ²
	7	5.66x10 ⁵	4.00x10 ²
	12	Too numerous	9.5x10 ³
Pasteurized + 0.04% sodium benzoate + 0.01% sodium azide	1	2.26x10 ⁴	1.96x10 ²
	7	6.70x10 ⁴	4.66x10 ²
	12	Too numerous	9.2x10 ⁴
Pasteurized + 0.02% sodium azide	1	7.6x10 ¹	1.3x10 ¹
	7	1.3x10 ¹	1.3x10 ¹
	12	3.1x10 ¹	2.1x10 ¹
Pasteurized tiger-nut milk without preservatives	1	2.03x10 ⁵	2.3x10 ³
	7	4.33x10 ⁶	7.86x10 ⁶
	12	Too numerous	1.2x10 ⁵

Each value is the mean of triplicate determinations

Table 4: Bacterial count of tiger-nut milk under storage conditions [40]

Treatments	Period of storage (week)	Microbial count (cfu/mL)	
		Total viable count	Fungal count
Refrigerating temperature			
Soaked tiger-nut beverage + Nisin preservative (2 g/ 2 L)	1	7.2x10 ⁵	7.6x10 ⁵
	2	7.8x10 ⁵	7.9x10 ⁵
	3	7.9x10 ⁵	8.4x10 ⁵
Soaked tiger-nut beverage	1	8.5x10 ⁵	8.7x10 ⁵
	2	8.5x10 ⁵	8.1x10 ⁵
	3	8.1x10 ⁵	8.2x10 ⁵
Room temperature			
Soaked tiger-nut beverage + Nisin preservative (2 g/ 2 L)	1	6.3x10 ⁵	8.0x10 ⁵
	2	7.7x10 ⁵	8.1x10 ⁵
	3	6.9x10 ⁵	7.8x10 ⁵
Soaked tiger-nut beverage	1	8.1x10 ⁵	8.2x10 ⁵
	2	8.4x10 ⁵	8.2x10 ⁵
	3	7.8x10 ⁵	8.6x10 ⁵

Values are means of triplicate determinations

Table 5: Bacterial load of tiger-nut milk under storage conditions ^[41]

Microbial count (log ₁₀ cfu/mL)	Storage period (day)	Storage condition	Tiger-nut milk				
			Control	+garlic	+ginger	+citric acid	+ginger +garlic
Total viable count	0	4°C	< 1	< 1	< 1	< 1	< 1
	4		1.40	< 1	1.37	< 1	1.09
	8		2.48	1.10	2.51	1.05	1.12
Total viable count	0	28°C	< 1	< 1	< 1	< 1	< 1
	4		1.71	1.22	1.62	1.00	1.11
	8		4.68	4.28	4.35	2.57	4.25
Fungal count	0	4°C	< 1	< 1	< 1	< 1	< 1
	4		1.25	< 1	< 1	< 1	< 1
	8		2.71	2.16	2.56	1.09	1.06
Fungal count	0	28°C	< 1	< 1	< 1	< 1	< 1
	4		1.53	1.34	1.58	1.03	1.38
	8		2.52	2.34	2.58	2.31	2.40

Each data is the mean+ standard error of 3 determinations

1.4 Bacteria isolated from milks storage at different temperature

Among bacteria isolated from tiger-nut milk under different temperature of storage (table 6), *Bacillus spp* are spore-forming bacteria that are commonly found in soil, water (through soil-water contamination) and on vegetables. *Bacillus spp* and *Aspergillus spp* isolated from the tiger-nut milk during storage has been isolated by other workers in pasteurized tiger-nut milk during storage ^[42] ^[43]. It was observed that the most predominantly encountered species were *E. coli* and *Bacillus spp*. which had 18.9% each ^[41]. The presence of these bacteria can be explained, because the spores of some strains of these organisms are resistant to pasteurization temperature. Others include *Enterococcus spp.* (16.2%), *S. aureus* and *P. aeruginosa* (13.5% each) and

Streptococcus spp. (10.8%). In spite of the use of phosphate buffer saline only and surface sterilized in 1% sodium hypochlorite for 5 minutes and then washed with phosphate buffer saline before extract the milk, different bacteria species isolated were made up of *Bacillus spp.* and *Escherichia coli* (18.9%) each, *Enterococcus spp.* (16.2%), *Pseudomonas aeruginosa* and *Staphylococcus aureus* (13.5%) each, *Streptococcus spp.* (10.8%) and 8.1% for *Enterobacter cloacae* ^[41]. Only *Lactobacillus spp.*, *Aspergillus niger* were isolated from tiger-nut milk during the 12 days period of ambient storage using the combination of heat treatment and 0.02% sodium azide ^[24]. Studies have reported that the use of sodium azide (0.02%), azidiol (0.4% of sodium azide and chloramphenicol) and bronopol (0.04%) can extend the shelf-life of raw milk samples up to 96 hours unrefrigerated ^[44].

Table 6: Bacteria isolated from tiger-nut milk at different conditions of storage ^[24] (1) ^[45] (2)

Treatments	Microorganisms
Ambient temperature (28±2°C)¹	
Pasteurized only	Bacillus spp, Lactobacillus spp, Pediococcus spp, Aspergillus niger, Aspergillus flavus, Aspergillus ochraceus
Pasteurized + 0.08% sodium benzoate	
Pasteurized + 0.04% sodium benzoate + 0.01% sodium azide	
Pasteurized + 0.02% sodium azide	Lactobacillus spp, Aspergillus niger
Ambient temperature (28 to 32°C)²	
Tigenut milk	Clostridium sordelli, Clostridium sporogenes, Staphylococcus schleiferi, Staphylococcus cohnii, Neisseria mucosa, Saccharomyces pombe
Tigenut milk treated + 0.05% sodium benzoate	Acinetobacter calcoaceticus, Staphylococcus cohnii, Corynebacterium xerosi, Vibrio parahaemolyticus, Saccharomyces pombe
Tigenut milk Irradiated + Ultraviolet Light	Micrococcus luteus, Saccharomyces pombe
High Temperature Short Time Pasteurized tigenut milk at 90 to 95°C for 15 to 30 seconds	Aeromonas veronii, Bacillus cereus, Bacillus alvei, Bacillus subtilis, Corynebacterium xerosis, Saccharomyces pombe, Saccharomyces cerevisiae
Refrigerating temperature (0°C)²	
Tigenut milk without treatment	Acinetobacter calcoaceticus, Serratia fonticola, Bacillus stearothermophilus, Clostridium sporogenes, Clostridium sordell, Saccharomyces pombe
Tigenut milk treated + 0.05% sodium benzoate	Clostridium sordelli, Serratia fonticola, Staphylococcus schleiferi, Staphylococcus simulans, Saccharomyces pombe

1.5 Sensory acceptability of milks added preservatives

Most preservative methods are used to extend the shelf-life of soymilk and tiger-nut milk. So, the combination of pasteurization and the addition of 0.02% sodium azide in tiger-nut milk was generally preferred by in all the sensory overall quality up to the twelfth day ^[24]. The tiger-nut drink

treated with ginger was preferred (4.1 - 4.7) in its overall quality acceptability throughout the storage period (refrigerated) and this was significantly higher than the value for the other samples on day 8 ^[28]. Sensory characteristics of tiger-nut milk are presented in table 7.

Table 7: Sensory quality of tiger-nut milk during ambient temperature (28°C) ⁽¹⁾ [24] ⁽²⁾ [41]

Treatments	Period of storage (day)	Score of overall acceptability	Acceptability
Pasteurized + 0.08% sodium benzoate ⁽¹⁾	1	1.2 ± 0.1	Like extremely
	7	2.6 ± 0.2	Like very much
	12	6.6 ± 0.2	Dislike very much
Pasteurized + 0.04% sodium benzoate + 0.01% sodium azide ⁽¹⁾	1	2.0 ± 0.2	Like very much
	7	4.6 ± 0.4	Neither like nor dislike
	12	6.7 ± 0.2	Dislike very much
Pasteurized + 0.02% sodium azide ⁽¹⁾	1	1.2 ± 0.1	Like extremely
	7	1.2 ± 0.1	
	12	1.2 ± 0.1	
Pasteurized tiger-nut milk without preservatives ⁽¹⁾	1	2.0 ± 0.2	Like very much
	7	6.5 ± 0.2	Dislike very much
	12	6.8 ± 0.1	Dislike very much
Tiger-nut extract (control) ⁽²⁾	0	4.6 ± 0.03	Good
	4	2.9 ± 0.06	Bad
	8	ND	-
Tiger-nut +garlic ⁽²⁾	0	4.0 ± 0.09	Good
	4	2.6 ± 0.09	Bad
	8	ND	-
Tiger-nut +ginger ⁽²⁾	0	4.1 ± 0.05	Good
	4	3.7 ± 0.08	Neither good nor bad
	8	ND	-
Tiger-nut +citric acid ⁽²⁾	0	4.3 ± 0.01	Good
	4	3.4 ± 0.03	Neither good nor bad
	8	ND	-
Tiger-nut +ginger +garlic ⁽²⁾	0	4.4 ± 0.01	Good
	4	2.7 ± 0.07	Bad
	8	ND	-

⁽¹⁾ Each value is the mean ± standard error of 10 member panelist. ⁽²⁾ Each data is the mean ± standard error of 12 member taste panelist.

1.6 Focus on soymilk and tiger-nut milk preservatives

Tiger-nut milk has a very short shelf-life of often less than 24 hours depending on the condition of storage [46]. High temperature and humidity significantly reduce the shelf-life of the product [47]. As a result, tiger-nut milk is often associated with significant microbial contamination, including bacteria and molds [47][48]. The short shelf-life of raw tiger-nut milk hinders widespread consumption of the beverage due to the deteriorating effects of some microorganisms on the milk [49]. In the literature, information on the extension of shelf-life of tiger-nut milk using combination of heat treatment with chemical preservatives is reported. The use of azidol to extend the shelf-life of fresh milk was reported. From the research of [23] it was reported that the shelf-life of cow milk was extended using azidol (sodium azide/chloramphenol) as the storage temperature was lowered from 20°C - 4°C for 3 days. The research reported the extension of the shelf-life of tiger-nut milk extract stored at 4°C for 8 days using combinations of heat treatment (80°C) and natural preservatives (citric acid, garlic, ginger, and clove) but with little success that could allow long distance transportation of the product and lengthened the storage time [28]. The use of flavouring agents such as ginger (*Zingiber officinale*) and garlic with 3% or 5% each other in the spicy lactic fermented tiger-nut milk drinks reduce total soluble solids content but it increase ethanol content [27]. Processing treatments has effect on the stability and acceptability of tiger-nut beverage products [40]. So, [45] reported the use of ultraviolet light and sterilization methods were more effective at eliminating most of the bacteria implicated in milk spoilage. Some work reported that the shelf-life of tiger-nut milk treated by combine heat treatment and sodium azide can extend up to 12 days of ambient storage [24].

Drying or dehydration helps to increase shelf-life and easier transportation and storage [50]. The soymilk can be kept for up to 13 days at refrigeration without no multiplication of mesophilic aerobes [25]. The sterilized soymilk at 121°C/ 15 min after bottling is acceptable up to 170 days in refrigerated condition (4°C) while same samples is acceptable up to 90 days in ambient temperature condition from the day of preparation [51]. Recently processing is developing to preserve the deterioration of soymilk. According to the stabilization of soymilk, many preservatives are used. The addition of 700-800 parts per million (ppm) of sodium benzoate, pasteurization and refrigeration while a combination of 175 ppm of sodium metabisulphite and 400 ppm of sodium benzoate can preserve soymilk for about 11 days [25]. Other studies reported that potassium-meta-bi-sulphite (KMS), Sodium-benzoate and potassium sorbate can be used individually or in a combination to preserve soymilk. The mixed preservatives can give shelf-life of 17 days but decreases the colour, flavor, taste and overall acceptability of soymilk [26]. Cloves used as a preservative increases the shelf life of soymilk by 2 days at room temperature and 8 days in refrigerated condition. Guinea pepper extends the shelf life by 1 day at room temperature and 6 days in refrigerated condition while combination of these extracts maintain longer shelf-life of 12 days in the refrigerated condition and 2 days at room temperature [21]. The fresh soymilk applied with the heat treatment at 100°C for 10 minutes, and nisin 0.004%, nipagin complex esters 0.008%, dehydroacetate 0.002% can be maintained under upper limit of the microbial count (100 cfu/mL) for more than 6 days at 25°C, and 11 days at 4°C [29]. From other research, it was reported that the shelf-life of soymilk based juice can be increased by adding 0.5 - 3.0 grams of the *Aframomum danielli* spice

powder to every 200ml of the blend ^[30]. The sensory properties of the soymilk increased with decrease in nutritional quality indicating that methods which increase sensory properties of soymilk by reducing its beany flavor have lower nutritional qualities ^[52]. From all research, it arise that all preservation technique gives best results if stored at refrigerated condition as compare to storage at room temperature ^[39].

Conclusion

The present review work concludes that the shelf-life of tiger-nut milk and soymilk during storage depends on the preservatives processing method used and the storage conditions. The most preservatives tested were the chemical or synthetic one. Unfortunately, after a long period of storage, the hedonic acceptability is not acceptable. Various changes during storage affect nutritional value and the sensory acceptability of milks. The changes may be microbial and fermentation. Preservatives used up to know to extend tiger-nut milk and soymilk shelf-life combines preservatives methods. The shelf-life of soymilk or tiger-nut milk increased by different preservatives if stored at refrigerated condition.

Authors' Contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

Conflict of Interest

Authors have declared that no competing interests exist.

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