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Preservation of Some Physico-Chemical Properties of Soymilk-Based Juice with *Aframomum Danielli* Spice Powder

A.O Dauda*, G.O. Adegoke

Department of Food Technology, University of Ibadan, Ibadan, Oyo State, Nigeria, West Africa
*Corresponding author: adegboladauda@yahoo.com

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Abstract The effect of *Aframomum danielli* powder on the shelf life of physico-chemical properties of soy-milk based juice was studied. In this paper, attempt was made to investigate the preservative effect of the powder of *Aframomum danielli*, a local spice, on the physico-chemical properties of blends of soymilk and juices from fruits and vegetable by prolonging its shelf life. This was done by adding 0.5-3.0grams of the spice powder to every 200ml of the blend, while the control samples had no spice. The juice quality was analyzed for sensory attributes and physico-chemical parameters over six month's period of storage at interval of four weeks. Sensory evaluation of the juices was judged for colour, flavour, taste and general acceptability, on a nine-point hedonic scale, varying from "dislike extremely" (score 1) to "like extremely" (score 9), according to the method of [39]. Ten (10) untrained panel members carried out the sensory evaluation. Infrared spectrophotometer was used to identify functional groups in the powder of the spice, *Aframomum danielli*, responsible for the stability of the juice. Differences were observed in the colour and aroma of treated samples in comparison with the control, though the samples were generally accepted. The percentage losses recorded for the total soluble solids, ascorbic acids and total sugars of treated samples were far lesser than those of control over the same condition and period of storage. Some of the active components of the spice identified are 4-amino-acetophenone, N,N-dimethyl-2-chloroacetoacetamide, 3-beta-acetoxy-5-etienic acid, 6,10-dimethylundeca-5,9,-diene2-one, Phenyl-3-buten and 4-Phenyl butanone. The samples treated with the *Aframomum danielli* powder were better preserved than the untreated ones (control).

Keywords: *Aframomum danielli powder, soymilk, juice, shelf life, active components, preservative*

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1. Introduction

Fruits and vegetables are among the most important foods of mankind as they are not only nutritive but are also indispensable for the maintenance of health. Fresh fruits and vegetables are an essential component of a healthy diet, able to decrease the risk of cardiovascular diseases and cancer [3]. Apart from their nutritional and sensory properties, fruits and vegetables are currently recognized as active and protective agents [31].

In the last years, their consumption has continued to grow rapidly, which was linked to the increased public awareness of their health benefits, even if it remains below the recommended daily intake in many countries, due to barriers such as complacency and lack of willpower to change the diet [35]. However, this new trend has also hiked the chances of outbreaks of food poisoning and food infection related to consumption of fresh fruits and uncooked salad [6]. Fruits and vegetables have been cultivated since ancient times [28]. Fruits are not only consumed fresh but also used to produce jam, jelly, stewed fruit, marmalade, syrup and several types of soft drinks. Citrus fruits have been reported to be rich in antioxidants compounds, particularly ascorbic acid and phenols [13,19,36]. Ascorbic acid is well known for its strong antioxidant activity [15], while phenolic compounds have been widely investigated and characterized for their anticancer properties [18,20,24].

Today's society is characterized by an increasing health consciousness and growing interest in the role of food for maintaining and improving human well-being and consumer health. Many studies have been done on the physical, chemical and nutritional properties of fruits, such as sweet cherry [28,33,34,44], Plum [14,16], wild plum [8], Orange [40], berries [27,22]. Various methods have equally been reported to have been used for the preservation of juices and shelf life evaluation equally reported by many scientists. Hot fill and aseptic methods were reported by [11] to be efficient in maintaining physico-chemical characteristics of juice up to twelve months. Clarified cashew apple juice along with tannase treatment stored at 4°C was stable for two months [9]. It was also reported by [21] that cashew apple juice preserved with aqueous extract (hot and cold) of *Aframomum danielli* showed reduction in the vitamin C contents and sugars after two weeks. Also, [5] used a combined method of preservation i.e drying of cashew apples with osmotic dehydration and observed that osmotic pretreatment was efficient in reducing water activity but not effective in protecting the juice from oxidation.

High Pressure (HP) processing of fruit and vegetable, which is another method of preservation, offers the chance of producing food of high quality, greater safety and increased shelf-life [7]. The main requirement this new technology must meet is to ensure product microbial safety while preserving sensory and nutritional characteristics to obtain products more similar to fresh foods [41]. High pressure processing (HPP) can enable ready to drink juice processors to produce innovative products with fresh-like, natural-like attributes and natural-looking colours which are all aspects valued by consumers nowadays [12]. Increasingly, consumers not only want food products to be of high (sensory) quality but also to deliver specific benefits in terms of health, safety and environmental quality [43].

In the last two decades or thereabout, food scientists have attempted to develop new technologies that improve the quality and quantity of products, while consumers have also become more critical on the use of synthetic additives to preserve food safety or enhance characteristics such as colour and flavour [10]. The growing demand for slightly processed products with the same guarantees of quality or better than those treated by traditional methods of preservation has urged researchers to focus most of their efforts on studying new ways of extending the shelf life of fresh produce.

Soymilk, another key component of the blend, (also called soyamilk, or soybean juice and sometimes referred to as soydrink/beverage) is a beverage made from soybeans (*Glycine max*). It is a stable emulsion of oil, water, and protein. Soymilk is produced by soaking dry soybeans and grinding with water in a Hammer mill before cooking at boiling point for about 20 minutes. Soymilk contains similar proportion of protein as cow's milk; around 3.5% and 2% fat, 2.9% carbohydrate, and 0.5% ash. They have been blended with other food products to produce other products, most likely, with better nutritional qualities. Milk-based products all contain actual milk (and/or soymilk) or different combinations of modified proteins. Most of the popular milk-based beverages are good sources of protein, containing 10 to 40 grams per serving.

In recent times, spice antioxidants have raised considerable interest among food scientists, manufacturers, and consumers because of their natural antioxidants [26]. Consumers are increasingly aware of the risk posed by synthetic antioxidants due to their high volatility and instability at elevated temperature, hence the need to shift focus to the use of natural antioxidants [1,29,30]. *Aframonium danielli*, a natural antioxidant and spice has shown its potential as a preservative in some food system [2,17].

The genus "*Aframomum*" has about 50 species spread over West and East Africa and the wetter parts of tropical Africa from Senegal and Ethiopia to Mozambique and Angola [25]. *Aframomum* species are well known for their fruits with aromatic seeds, of which *Aframomum danielli* is the most common. *Aframomum danielli* tree is a large robust perennial plant (3-4m tall) found in Central and West African countries. It belongs to the family Zingiberaceae. It is known as "Oloburo" or "atare aja" in Yoruba speaking regions of Nigeria. The pods are usually brown in their dried state. The seeds are oval (about 2-3mm in diameter), olive brown and shiny in appearance and are commonly used for traditional dishes, especially pepper soup. The essential oil is used in dye preparations and flavorings.

In this paper, the aim is to give an insight into the knowledge or the use of *Aframomum danielli*, a local spice, in ensuring the physico-chemical stability and quality of soymilk-based juice.

2. Materials and Methods

Aframomum danielli pods were purchased from a local market in Ibadan, Oyo State, Nigeria. The seeds were removed from the pods and cleaned of the extraneous materials. The seeds were winnowed and milled into powder using hammer mill. The powder was then sieved with a wire-mesh to obtain fine powder and stored at room temperature for use. *Aframomum danielli* powder (0.5g – 3.0g) was added to every 200ml of the juice blends.

2.1. Juice Preparation

Fresh oranges were collected from a local farm at Ajibode village in Ibadan. Carrot and watermelon were purchased from Sabo in Ibadan, while the soybeans used for soymilk was obtained from International Institute for Tropical Agriculture (IITA) in Ibadan. These fresh, juicy, good quality fruits and vegetable were sorted for processing. The fruits and vegetable were weighed, washed thoroughly under tap water to remove foreign materials. Juices were extracted, filtered through sterilized muslin cloth with an average yield of 620ml/kg of each fruit and vegetable used. The soybeans were cleaned properly, soaked in water at (30^{o}C for 5hours), grinded (with hammer mill), slurry cooked (at boiling point for 20 minutes), separation of the cooked soymilk from fibre and boiling of the resulting filtrate to obtain soymilk. The laboratory prepared soymilk, juice from the vegetable and fruits were blended in equal ratios. Every 200ml of the blend was treated with powder of *Aframomum danielli* (0.5g – 3.0g).

2.2. Shelf Life Study

The shelf life study of the soymilk-based juice was based on sensory and physico-chemical quality of the juice products.

2.3. Sensory Analysis

Sensory evaluation of the juices was carried out on the colour, flavour, taste and general acceptability using a nine-point hedonic scale, varying from "dislike extremely" (score 1) to "like extremely" (score 9) according to a method of [39]. 10 untrained panel members carried out the sensory evaluation. The juices were served in a coded and transparent white glass cups for proper assessment. The juices were served in triplicate and coded separately.

3. Physico-Chemical Analyses

pH of the juice was determined by weighing 10 grams (v/w) of the test portions into a clean Erlenmeyer. The decanted supernatant in a 250ml beaker was used to determine the pH using electrode and pentiometer standardized with buffer solutions of pH 9.18 and 4.01 at temperature 25^{0}C and expressed as pH units.

Total Soluble Solids (TSS) was determined using a hand digital refractometer (Atago, Tokyo model Leica 10431) according to the method of [45].

Total Titratable Acidity was determined by titrating 10ml of the extract (juice) with 0.1N Na0H after 1ml of the sample was diluted into 100ml volumetric flask, 1% phenolphthalein added and shaked properly. It was then titrated.

Vitamin C content (in mg/100ml) was determined using High Performance Liquid Chromatography (HPLC).

Total sugar from the juice was done according to the method of analysis by [45].

Statistical Analysis: Data were analysed using (ANOVA) at P = 0.05.

4. Result and Discussion

Sensory Analysis: There were significant differences in the colour and aroma of the samples. After the period of storage, it was noticed that the colour which was slightly cloudy (treated samples) began to depreciate. The hedonic scale rating of both the untreated (represented by CTR) and treated samples showed that they were accepted. There was no significant difference in the overall acceptability of the samples over the period of storage, though the colour experienced little change (Table 1). The changes experienced in the colour of the samples could be due to the elimination of coloured pigments (carotenoids and others) from the fresh juice. After three and half months of storage, the juice was not acceptable.

Table 1. Sensory Evaluation (mean hedonic scores) of the Treated and Untreated Samples of COWS

Sample	Colour /Appearance	Taste	Aroma /Flavour	Overall acceptability
CTR	6.73^a	6.27^a	6.07^{bc}	6.17^a
A	6.25^{ab}	6.33^a	5.83^{ab}	6.20^a
B	6.25^{ab}	6.27^a	5.57^b	6.33^a
C	5.72^b	6.00^a	5.37^c	6.20^a
D	5.03^c	5.80^a	5.37^c	6.03^a
E	5.13^{bc}	6.03^a	5.80^{ab}	6.03^a
F	5.73^b	6.17^a	6.37^a	6.20^a

Superscripts down a column are significantly difference with a>b>c>d.
Means for each attributes followed by the same letter are not significantly different at 5% level by Tukey.

CTR = Control samples (untreated Samples)
A = Juice blend treated with 3 grams of *Aframomum danielli* powder
B = Juice blend treated with 2.5 grams of *Aframomum danielli* powder
C = Juice blend treated with 2.0 grams of *Aframomum danielli* powder
D = Juice blend treated with 1.5 grams of *Aframomum danielli* powder
E = Juice blend treated with 1.0 grams of *Aframomum danielli* powder
F = Juice blend treated with 0.5 grams of *Aframomum danielli* powder
COWS = Blends of Carrot, Oranges, Watermelon and Soymilk

5. Physico-Chemical Analyses

The results are as shown in Table 2 to Table 6. The pH values of the preserved juice samples decreased from 4.97-3.88 (Table 2) over the period of storage (180 days). The decrease could be attributed to the growth of microorganism that produced lactic acid. Most bacteria will not grow at low pH and hence good keeping quality of the juice was maintained.

Table 2. pH of Blends of Carrot, Oranges, Watermelon and Soymilk Treatment with *Aframomum danielli* Powder (in grams)

Storage Time (in weeks)	3g	2.5g	2.0g	1.5g	1.0g	0.5g	CTR
0	4.97a	4.93a	4.66a	4.89a	4.95a	4.88a	4.91a
4	4.95a	4.79b	4.55b	4.83	4.91a	4.76b	4.82b
8	4.89b	4.61c	4.53b	4.74b	4.77b	4.67c	4.63c
12	4.84c	4.52d	4.46c	4.46c	4.60c	4.51d	4.52d
16	4.78d	4.38e	4.40c	4.40c	4.48d	4.37e	4.44e
20	4.68e	4.19f	4.07d	4.07d	4.32e	4.22f	3.97f
24	4.61f	3.93g	3.88e	3.88e	4.18f	4.16g	3.75g

Means for each attributes followed by the same letter are not significantly different at 5% level by Tukey.

Total Soluble Solids (TSS) – Total Soluble Solids of the juices indicate maturity of fruits used as reported by [42]. Total soluble solids of the treated samples were in the range of 42.83mg/L- 49.93mg/L over 180days storage period, while the control samples were between 15.62mg/L-49.93mg/L over the same period (Table 3). The stability of the total soluble solids of the juice indicates that the fruits were matured and could be responsible for good palatability and acceptability of the juice samples and could equally be attributed to the effects of the preservatives used. The slight decrease of TSS in the treated samples could be attributed to the effect of the preservative added, while larger decrease in the untreated samples might be due to the utilization of sugars by fermenting organisms leading to sugar degradation.

Total titratable acidity of the Samples was in the range of 0.14%-0.50% (Table 4) as malic acid present in the juice and was stable for over 14weeks of storage. The stability of the acidity indicates the concentration of organic acids mainly malic acid present in the juice to be stable. The decrease noticed in the titratable acidity might be due to the release of acids by decomposition, hydrolysis, oxidation or fermentation, which modifies the hydrogen ion concentration and consequently, food acidity. There were significant differences in the titratable acidity of the treated and untreated samples. There was increase in the titratable acidity of both the treated and untreated samples. The observed differences in the increase of the titratable acidity of both the treated and untreated samples could be attributed to the effect of the preservative used. Increase in titratable acidity of untreated samples compared to the treated ones showed that chemical degradation has been prevented in treated samples by the preservatives.

Table 3. Blends of Carrot, Oranges, Watermelon and Soymilk (mg/L Total Soluble Solids) Treatment with *Aframomum danielli* Powder (in grams)

Storage Time (in weeks)	3g	2.5g	2.0g	1.5g	1.0g	0.5g	CTR
0	49.93a	49.93a	49.93a	49.93a	49.93a	49.93a	49.93a
4	49.81a	48.40b	48.39a	49.92a	48.61b	46.43b	48.92b
8	49.32a	45.51c	43.44c	48.91b	48.72b	47.52b	49.63a
12	46.21b	43.74d	35.09d	45.53c	47.90c	46.23c	29.72c
16	44.81c	39.82e	29.19e	42.69d	47.86c	43.53d	18.31d
20	43.21c	34.96f	28.75e	38.26e	42.68e	37.92e	17.44d
24	28.30d	28.03g	27.93f	28.62f	26.91e	26.83f	15.62e
% loss	43.32	43.86	44.06	42.68	46.10	46.26	68.72

Means for each attributes followed by the same letter are not significantly different at 5% level by Tukey.

Table 4. Blends of Carrot, Oranges, Watermelon and Soymilk (% TTA) Treatment with *Aframomum danielli* Powder (in grams)

Storage Time (in weeks)	3.0g	2.5g	2.0g	1.5g	1.0g	0.5g	CTR
0	0.14f	0.08e	0.14f	0.08e	0.12g	0.16	0.04e
4	0.21e	0.12d	0.18e	0.11d	0.14d	0.18e	0.07e
8	0.28d	0.14d	0.20e	0.16c	0.19e	0.23d	0.10d
12	0.35c	0.16c	0.27d	0.21b	0.24d	0.27c	0.15c
16	0.40b	0.18c	0.38c	0.24b	0.29c	0.34b	0.18c
20	0.51a	0.27b	0.47b	0.32a	0.33b	0.36b	0.27b
24	0.50a	0.34a	0.50a	0.34a	0.39a	0.44a	0.57a

Means for each attributes followed by the same letter are not significantly different at 5% level by Tukey.

Ascorbic acid is an index of nutrient quality of fruits and vegetable and much more sensitive to various modes of degradation in food processing and subsequent storage [32,37]. Ascorbic acid is also used as a standard for monitoring the quality of juices in storage. Vitamins C content of the juice was 0.64 ± 0.0mg/ml and was stable for over 12weeks before slight degradation started. Between 8%-13% degradation was observed in the treated samples over the storage period, while untreated sample had over 35% degradation for the same period of storage (Table 5).

Table 5. Blends of Carrot, Oranges, Watermelon and Soymilk (Vit C in mg/ml) Treatment with *Aframomum danielli* Powder (in grams)

Storage Time (in weeks)	3g	2.5g	2.0g	1.5g	1.0g	0.5g	CTR
0	0.64a	0.64a	0.64a	0.64a	0.64a	0.64a	0.64a
4	0.61a	0.63a	0.63a	0.62a	0.64a	0.64a	0.57b
8	0.61a	0.62a	0.61a	0.63a	0.63a	0.62a	0.51c
12	0.60a	0.60a	0.61a	0.61a	0.63a	0.60c	0.44d
16	0.57b	0.60a	0.61a	0.60a	0.62a	0.60b	0.44d
20	0.57b	0.59b	0.60a	0.59b	0.61a	0.59b	0.41d
24	0.56b	0.58ba	0.59b	0.58b	0.59b	0.58b	0.40e
% loss	12.75	9.49	8.71	9.33	7.62	9.18	38.41

Means for each attributes followed by the same letter are not significantly different at 5% level by Tukey.

Higher losses in the untreated samples are probably due to the absence of preservatives. The decrease observed could also be attributed to oxidation, which occurs in fruit juices during storage and is highly dependent on the presence of oxygen in the head space or dissolve in the juice [11]. Some other researchers have reported decrease of Vitamin C in stored juices. Concord grape juice preserved with potassium sorbate and stored in glass bottles at 37°C retained much of its ascorbic acid for five months [38] and orange juice packed in glass bottles was reported to maintain ascorbic acid concentration for 112 days at 22°C [4]. It was also reported by [11] that 25.65% decrease was observed for hot fill and 26.74% for aseptic method after 350days of storage. According to [23], orange juice should contain at least 25mg vitamin C per 100ml at the time of expiration, which is about 50% of initial amount. Ascorbic acid loss of between 8 and 13% in the *Aframomum danielli* treated samples indicated that the preservative helped in preventing degradation and assist in the retention of over half of the ascorbic acid contained initially.

Total sugars (Table 6) were stable over the period of storage. Between 1%-5% of total sugars were lost in the treated samples after 24 weeks of storage, while untreated samples lost over 8% of its sugars over the same period. The loss recorded might be due to non-enzymatic browning reactions, either caramelization or maillard reactions occurring between amino acids or reducing sugars. Non enzymatic browning during processing or storage of juices may affect the flavour, colour, or other quality factors of the product. It is the most complex reaction in food chemistry due to the large number of food components able to participate in the reaction through different part ways giving rise to a complex mixture of products.

Table 6. Blends of Carrot, Oranges, Watermelon and Soymilk (% Sugar) Treatment with *Aframomum danielli* Powder (in grams)

Storage Time (in weeks)	3g	2.5g	2.0g	1.5g	1.0g	0.5g	CTR
0	19.61a	19.61a	19.61a	19.61a	19.61a	19.61a	19.61a
4	19.51b	19.60a	19.52b	19.50b	19.50b	19.42c	18.82b
8	19.30c	19.49b	19.45c	19.46bc	19.53b	19.52b	18.43d
12	19.23d	19.50b	19.41d	19.48bc	19.54b	19.50b	18.54c
16	19.20d	19.31c	18.88c	19.39c	19.21c	19.40c	18.27c
20	19.09de	19.20d	18.72f	19.28d	19.03d	19.38cd	18.12f
24	18.99e	19.14e	18.53g	19.25d	18.93e	19.21d	17.90g
% loss	3.16	2.40	5.51	1.84	3.47	2.04	8.72

Means for each attributes followed by the same letter are not significantly different at 5% level by Tukey.

6. Conclusion

The results of the experiment confirmed that *Aframomum danielli* powder is a good natural additive for the preservation of the physico-chemical properties of soymilk-based juice. It effectively retained the nutritional qualities of the juice, as attested to by the stability of total soluble solids, vitamin C and total sugar contents of the juice. Sensory attributes of the juice was accepted. The local spice, *Aframomum danielli*, utilized in the research work is cheap, available and has no recorded side effect. The use of *Aframomum danielli* powder in the preservation of juice products and other food should be encouraged.

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