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PRESERVATIVE EFFECTS OF *GMELINA ARBOREA* FRUITS AND *NAUCLEA LATIFOLIA* STEM BARK EXTRACTS ON FRUIT JUICE IN COMPARISON WITH A KNOWN CHEMICAL PRESERVATIVE

– Research paper –

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Abstract: Fruit juices are liable to spoilage as a result of fermentation by microorganisms. This study is intended to provide information on preservative of fruit juices with plant extracts. The preservative effects of *Gmelina arborea* fruit and *Nauclea latifolia* stem bark extracts in apple and pineapple juices were assessed in comparison to chemical preservative (sodium benzoate) as a positive control and refrigeration at 4 ^{o}C as a negative control measures. Decrease in microbial load during storage was observed in the prepared juice samples. *G. arborea* fruit extract in microbial inhibition was more potent followed by sodium benzoate and *N. latifolia* stem bark extract. *G. arborea* preserved juices were of better choice in sensory evaluation for acceptability than *N. latifolia* and sodium benzoate preserved juices. Phytochemical screened from the extracts are saponins, tannins, flavonoids, alkaloids and steroids. The titratable acidity of the prepared juices evaluated *G. arborea* of lower titratable acidity value than *N. latifolia*. The results has provided a partial support for the use of *G. arborea* fruits and *N. latifolia* stem bark extracts for preservation of fruit juices. The use of *N. latifolia* and *G. arborea* as preservative agents have not been documented and could be potential sources of natural preservative agents for future use in preservation of alcoholic and non alcoholic beverages.

Keywords: Fruit juice, Preservation, Microorganisms**,** Plant extracts

INTRODUCTION

Plant extracts could serve as preservatives as does by chemical preservativesin extending the shelf life and maintainance of quality in fruit juices. The demand for nutritious foods such as fresh fruits and fruits crush not pasterized by consumers have escalated in the recent time owing to high amount of ascorbic acid, low contents of salt and other vital natural substances which are so much important in heart diseases prevention and also in cancer and diabetes prevention (Matthew, 2006; Kumar et al., 2009; Patrignaniet al., 2010; Ginter and Simko, 2012). Fruits benefits in health care and their availability are reduced as a result of microbial spoilage. Several emerging spoilage microorganisms are of great concern in fruit juice industries; for example, *Alicyclobacillus acidoterrestris* has been isolated from several fruit drinks and fruit products with infection rate that ranged from 14.7% - 18.3%. *Propionibacterium cyclohexanicum* and those imperfect fungi having heat resistantproperties as found in *Talaromyces trachyspermus*, *Neosartorya fischeri*, *Byssochlamys nivae* and *Byssochlamys fulva* have also been implicated in fruit juices spoilage (Walker and Phillips, 2007; Steyn et al., 2011). For prevention of these microorganisms in fruit juices, thermal treatment is the effective method for microbial inactivation but it may produce unwanted characteristics on foods like nutrient loss or also freshness reduction like flavor (Kuldiloke at el., 2008; Carbo et al., 2010). Chemical preservatives, such as benzoic acid and potassium (2E, 4E)-2, 4-hexadienoate (Potassium sorbate) are commonly employed in fruit juices and beverages to extend their shelf life (Walker and Phillips, 2008). However, consumers demand for safe and fresh foods which are not preserved with chemicals, leads to the increased rate forusing preservatives derived from naturein foods (Raybaudi et al., 2009). Natural preservatives as found with bacteriocins from lactic acid bacteria, plants derived essential oils, chitosan from the skeleton of crabs, lobster and shellfish, organic compounds such as sorbic,

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lactic and propionic; and food phenolic compounds found in vegetables, beverages and plants have all received credibility in food preservation (Rico et al., 2007; Raybaudi et al., 2009; Aneja et al., 2014).

Apple is a popular known fruit and it is consumed all over the world(Potter et al., 2007). Apples have health benefits as it isrich in antioxidants (Lee et al., 2003; Boyer and Liu, 2004), plant nutrients and some minerals essential for cell growth and body development. Pineapple (*Ananascomosus*) is a member of the tropical plants (bromeliads) which in that family can only be eaten. Pineapple has inherent proteolytic enzymes that is used to aid digestion and exogenous proteolytic enzymes to enhance meat tenderness (Cheesbrough 2000).

Recentlysome researchers have suggested natural preservatives to improve fruits and fruit products to replace chemical preservatives (Jeong et al., 2008; Krzystof et al., 2010).This study analyses two candidate's natural preservative sources from *G. arborea*and *N. latifolia*, compared with the preservative effect of a chemical preservative (sodium benzoate) as potential fruit juice shelf lifeextender.

MATERIALS AND METHODS

Sample collection: Fresh apples and pineapples were obtained in sterile nylon bags from a local market in Ado-Ekiti, Ekiti State, Nigeria. The plant extract (*Nauclea latifolia*) stem bark was scraped off from the tree at Erifun village, close to Afe Babalola University, Ado Ekiti. Healthy looking matured fruits of *Gmelina arborea* were picked underneath *G. arborea* tree at Afe Babalola University.

Plant extracts preparation: Matured *G.arborea*fruits were obtained, soaked in soap solution for two minutes and washed. The washed fruits were then rinsed severally with distilled water. The seeds were removed and the mesocarp was again rinsed severally with distilled water. After which, 100 grammes was weighed and homogenized with Malex blender (model M-002nv). The obtained juice was filtered through triple layered clean mousseline and passed through filter paper (Whatman number 1) to obtain impurity free extract and finally, through membrane filter for sterility. Before use, the extract was stored in a sterile brown sampling bottle and stored at room temperature (28±2 ^{o}C). *Nauclea latifolia* extract was prepared by scraping the stem bark from the tree, washed thoroughly and rinsed in clean water. After which it was shed dried in the laboratory for 14 days and was pulverized to smooth powder with a grinding machine. One kilogramme (kg) was obtained and dissolved inethanol (500 ml) for 24 h. It was passed through filter paper (Whatman number 1). The filtered extract was evaporated with rotary evaporator (RE -52 A Union Laboratories, England) at 45 ^{o}C to obtain semi solid extract. This extract was kept in a brown sterile bottle and stored at room temperature (28±2 ^{o}C) before use.

Sterility test: Sterility test of extracts was performed by streaking a loop full of each extract on freshly prepared plates of Nutrient agar (NA) and Potatato Dextrose agar (PDA). The nutrient agar plates for bacterial growth were incubated for 24 – 48 h, while the potato dextrose agar plates for fungal cultivation were incubated for 72 h at 28±2 ^{o}C. Absence of microbial growth on streaked lines after periods of incubation approved sterility of the extracts.

Antimicrobial test: Well-in-agar method was employedto determine antimicrobial activities of the extracts. One gramme of *N. latifolia* extract was reconstituted with 10 millilitres of sterile distilled water while *G. arborea* fruit juice was used without reconstitution. Mueller Hinton Agar culture plates and Potato Dextrose agar plates were inoculated with 10^{-7} CFU of the bacteriaand 10^{-7} spore/ml of fungi species to be tested for susceptibility and were stand to solidify; and seeded microorganisms established in the media. With a cork borer size of 4 mm, wells were made in the gelled agar. Using a micro pipette, 0.5 ml of extracts were filled into each well.The bacterial cultured plates were incubated for 24 h at 37 ^{o}Cwhile fungal cultured plates on potato dextrose agar were incubated for 74 h at 28±2 ^{o}C.Inhibition zones were measured at end on incubation and reported against the tested microorganisms.

Production of apple and pineapple juice: Fruits were washed with soap solution and rinsed severally with distilled water to remove

traces of soap. The fruits were peeled with clean and sharp knife, specks removed and diced. The diced apple and pineapple were homogenized with a warring electric blender separately and the juice extracted was filtered by passing through sterile triple layered moussline to remove suspended materials and finally through a sterile filter of 0.2 mm pores size. Four hundred millilitres of each juice was dispensed aseptically into four sterile bottles and were simmered for 5 minutes in water bath regulated at 80 ^{o}C.They were removed from water bath and allowed to cool. One bottle each of apple and pineapple juice were separately preserved with 1 mg/ml concentration of *G. arborea* fruit extract, 1 mg/ml of *N. latifolia* stem bark extract, sodium benzoate (positive control) and the fourth set of apple and pineapple juice without preservative (negative control) in refrigerator. Both the chemically and extracts preserved juices were stored at room temperature while unpreserved set of juices were refrigerated at 4^{o}C.

I*solation, characterization and identification of bacteria and fungi isolates:* An aliquot of the apple and pineapple juice was serially diluted into 10^{-5} dilutions using sterile distilled water and 1 ml of 10^{-4} dilution was pour plated on nutrient agar plate and 1 ml of 10^{-3} on PDA plates to isolateassociated bacteria and fungi species respectively from each of the fruit juice before pasteurization. The bacterial growth plates were incubated for 24 h at 37 ^{o}C and fungal growth plates at room temperature (28±2 ^{o}C) for 72 hours. Also after pasteurization, at days 0, 5 and 10, 1ml of each juice sample was obtained aseptically and serially diluted and plated as did for unpasteurized samples for bacterial and fungal growth. Using colony counter, resultant bacterial colonies were enumerated and distinct colonies from culture plates were purified by sub-culturing and obtained pure cultures were transferred to agar slants and stored in refrigerator (4 ^{o}C) for characterization and identification.

The bacterial isolates were identified culturally, morphologically and biochemically according to the criteria of Holt et al.(1994); Sneath et al.(1986).

Two drops of lacto phenol in cotton blue solution was dispensed on mycelia mat directly on plates to avoid disruption of the fungi natural structures. The mycelia mat was then observed under low power and medium objectives of microscope. Base on the criteria of (Barnett et al., 2000) the fungi isolates were identified to species level.

Extracts phytochemical analyses

Chemical methods of testing for the presence of phytochemicals such as alkaloids, saponnins,flavonoids, tannins and steroids were carried out with the criteria of (Trease and Evans, 1989; Harbone and Williams, 2000).

Sensory evaluation of fruit juice: Equal volume of 150 ml each juice sample was dispensed into a transparent glass cup to evaluate sensory parameters with a 10 member panel of regular juice drinkers. The sensory quality evaluated include: appearance, color, flowing properties, aroma, flavour, taste, texture, thickness, mouth full and overall acceptability. The parameters rated on a 9 point scale were 1 (dislike slightly), 2 (dislike moderately), 3 (dislike very much),4(dislike extremely), 5 (neither likenor dislike), 6 (like slightly), 7 (like moderately) 8 (like very much) and 9(like extremely). This experiment was repeated 4 times to re-taste and change their scores if necessary. At interval, clean water was supplied to rinse their mouth before each taste. The data obtained were subjected to analysis of varianve (ANOVA) and Ducan's multiple range test was used for separation of mean.

Titratable acidity: 25 ml of juice sample was poured in a beaker and two drops of phenolphthalein as indicator was added. This was titrated with 0.1 Nornal sodium hydroxide (NaOH) until pink colour was reached. Reseults were reported as tartaric acid in percentage.

Statistical analysis: Results obtained were expressed as the mean ± S.E.M of triplicates. SPSS 10.0 for window soft wear package and Student's t-test for statistical analyses was used. Values were considered to be statistically significant at (P>0.05)

RESULTS AND DISCUSSION

Inhibition potentials of the extracts

The antibacterial activity of the extracts showed varied degree of inhibition. All tested bacteria species were susceptible to *G. arborea*extract, with *Staphylococcus aureus* being the most inhibited with zone of 31.3 mm followed by *Pseudomonas aeruginosa* with zone of 25.3 mm and least inhibition zone of 18.7 mm on *Klebsiella pneumoniae*. Among the fungi species, *Aspergillus flavus*was the most inhibited with a zone of 26 mm, followed by *Trichoderma viride* with a zone of 17.3 mm and least inhibited *Aspergillus niger* with a zone of 14 mm. Akyala et al., (2013)in ealier study have investigated the fruit extract of *G. arborea* antimicrobial potency on some pathogens such as *Pseudomonas aeruginosa, Staphylococcus aureus* and *Escherichia coli*. Among these isolates, *Staphylococcus aureus* and *Aspergillus niger* known as pathogenic organisms were isolated from the juice before pasteurization. However, spoilage organisms such as *Micrococcus luteus, Aeromonas hydrophila, Lactobacillus coryneformis* and yeast species were isolated after 5 days of storage.

The ethanol extract of *N. latifolia* inhibited *Bacillus cereus, Klebsiella pneumoniae, Pseudomonas aeruginosa* and *Proteus mirabilis* with varying susceptibility degree. *Bacillus cereus* was the most inhibited bacteria with a zone of 19 mm. This was followed by *Pseudomonas aeruginosa* with17 mm and *Klebsiella pneumoniae* with 16 mm, *Proteus mirabilis* was the least inhibited with a zone of 14.7 mm. Other tested microbes were resistant to this extract (Table 1). (Okeiei et al., 2011; Anowi et al., 2012), have reported*N. latifolia* extracts in varied degrees on *B. cereus, S. aureus, E. coli, C. albicans, P. aeruginosa,K. pneumoniae* and *A. niger*. In the study of (Khan et al., 2011), *Escherichia coli* and *Shigelladysentereae* were resistant to aqueous and N-Hexane extract of *N. latifolia*, with *Staphylococcus aureus* showing resistance only to the N-Hexane extracts.

This study on the antimicrobial of two candidate's natural preservative sources from *G. arborea*and *N. latifolia*, has helped to suggest the use of the plant extracts as potent plants to be employ for extendingfruit juice shelf life as the result attained can be compared with the preservative effect of a chemical preservative (sodium benzoate).

Table 1. Inhibition zone (mm) created by extract on test microorganisms

Test microbes	*Gmelina arborea*	*Nauclea latifolia*
Bacillus cereus	24±0.4	19±0.4
Enteroccus cloacea	21.7±0.4	-
Proteus mirabilis	24±0.0	14.7±0.4
Escherichia coli	20.3±0.4	-
Pseudomonas aeruginosa	25.3±0.4	17±0.2
Klebsiella pneumoniae	18.7±0.4	16.7±0.5
Staphylococcus aureus	31.3±0.7	
Aspergillus fumigatus	22.7±0.4	
Aspergillus niger	14±0.0	
Aspergillus flavus	26±0.6	
Trichoderma viride	17.3±0.4	

Phytochemicals screened from the plants

Qualitatively determined phytochemicals from *G. arborea* are saponins, alkaloids, flavonoids, steroids and phenol while *N. latifolia* contained saponins, tannins, alkaloids and phenol (Table 2). From the perspective of screened phytochemicals, the bioactive compounds such as saponins, phenol and flavonoids from *G. aborea* will desire it a good inhibitory strength for antimicrobial application than *N. latifolia* extract. Akyala et al. (2013), have also confirmed the presence of saponins, flavonoids and steroids in fruit of *G. arborea*. Maitera et al.(2011), have confirmed the presence of saponin, alkaloids and tannins in *N. latifolia*. These chemicals could demonstrate the inhibition of microbes from the preserved juices. Saponins have the activity to precipitate and coagulate red blood cells within injuries (Okwu and Okwu, 2010). Flavonoids provide anti-inflammatory and antifungal activity. Tannins having high potential antimicrobial properties have been used to hasten the healing of wound and inflamed mucous membranes (Egbung et al., 2011). Alkaloids poseseses anti-malaria activityas reported by Abbah et al. (2010); Odeyet al. (2012). Therefore, in conjunction with their preservative effects in juice, the extracts could help against inflammation and peptic ulcer and more health benefits than chemical preservatives as limitations have been reported on them.

Table 2. Phytochemical compositions of *Nauclea latifolia* and *Gmelina arborea*

Extracts	*Nauclea latifolia*	*Gmelina arborea*
Saponins	+	+
Tannins	+	-
Flavonoids	-	+
Alkaloids	+	-
Steroids	-	+

Preservative potentials of the plant extracts on juice samples

The extracts were observed to be of potential preservative agents as off flavour, sour and flat tastes were not observed in the preserved juices after two weeks. Before pasteurization, bacterial load of 66×10^4 cfu/ml was recorded from apple juice and 99×10^4 cfu/ml from pineapple juice. Fungal load of 54×10^3 spore/ml was recorded from apple juice and 63×10^3 spore/ml from pineapple juice.

After pasteurization, bacterial load of 25×10^4 cfu/ml and 42×10^4 cfu/ml; and fungal load of 20×10^3 spore/ml and 39×10^3 spore/ml were observed respectively from apple and pineapple juices. The plants' extracts preserved juice for 10 days storage at room and refrigerated temperature had varied microbial load. The *G. arborea* extract preserved juice had less microbial load compared to *N. latifolia* extract and sodium benzoate preserved juices. From the juice without preservative (control) but stored in the refrigerator, increase in microbial load was obtained from day 0 to 10^{th} day of storage. The highest microbial load recorded from apple juices preserved with *G. arborea*at day 0 was 25×10^4 cfu/ml and decreased to 5×10^4 cfu/ml at day 10 of storage. Fungal load of 20×10^3 spore/ml was observed at day 0 but decreased to 7×10^3 spore/ml at day 10 of storage. The bacterial load of pineapple juice preserved with *G. arborea* extract at day 0 was 42×10^4 cfu/ml and decreased to 3×10^5 cfu/ml at day 10 of storage. The fungal load at day 0 was 39×10^3 spore/ml and but decreased to 9×10^3 spore/ml at 10 day of storage.

Highest bacterial load recorded from apple juice preserved with *N. latifolia* extract at day 0 was 25×10^4 cfu/ml and decreased to 11×10^5 cfu/ml at 10 day of storage, while fungal load of 20×10^3 spore/ml at day 0 decreased to 10×10^3 spore/ml at 10 day of storage. The pineapple juice preserved with *N. latifolia* extractat day 0 has bacterial load of 42×10^4 cfu/ml but decreased to 14×10^5 cfu/ml at day 10 of storage and 39×10^3 spore/ml of fungal load at day 0 also decreased to 12×10^3 spore/ml at 10 day of storage.

Apple juice preserved with sodium benzoate had bacterial load of 25×10^4 cfu/ml at day 0 which decreased to 6×10^5 cfu/ml at day 10 of storage and 20×10^3 spore/ml of fungal load at day 0 which decreased to 5×10^3 spore/ml at day 10 of storage. The pineapple juice preserved with sodium benzoate at day 0 had bacterial load of 42×10^4 cfu/ml but decreased to 6×10^5 cfu/ml at day 10 of storage; and 39×10^3 spore/ml of fungal load decreased to 8×10^3 spore/ml at 10 day of storage.

The bacterial load recorded from apple juice with no preservative at day 0 was 25×10^4 cfu/ml and decreased to 10×10^5 cfu/ml at day 5 of storage but after which, increased to 18×10^5 cfu/mlat day 10. Fungal load of 20×10^3 spore/ml was observed at day 0 but decreased to 10×10^3 spore/ml at day 5 of storage and onday 10, increased to 15×10^3 spore/ml.

Pineapple juice with no preservative had bacterial load of 42×10^4 cfu/ml at day 0 and decreased to 10×10^5 cfu/ml at day 5 of storage, but however increased to 16×10^5 cfu/ml at day 10 of storage. Fungal load of 39×10^3 spore/ml observed at day 0 also decreased to 11×10^3 spore/ml at day 5 of storage and then increased to 19×10^3 spore/ml at 10 day of storage (Table 3).

Isolated microorganisms

From the preserved fruit juices, few organisms were isolated (5 bacteria and 6 fungi). The bacteria species isolated from preserved fruit juice were *Micrococcus luteus, Lactobacillus coryneformis, Zymomonas mobilis, Aeromonas hydrophilia* and *Staphylococcus aureus.* Isolated moold/yeast were *Penicillium italicum, Aspergillus niger*, *Candida krusei, Kleockera apiculata, Metschnikowia pulcharina* and *Schizosaccharomycespombe*. The isolated bacteria and fungi species are considered as spoilage microorganisms. The presence of yeast in the juice samples was expected due to its proliferation in samples with high sugar contents and low pH. The isolated bacteria species from the juice samples before preservation have been reported as the common spoilage organisms of wine due to low pH and this implies that the low pH level of the juice supported the growth of these organisms. Similar observation was recorded by Bevilacqua et al. (2011). Species of the genus *Lactobacillus* is one of the bacteria isolated from the juice. This species of bacteria

is common in animal feeds,milk and milkproducts, manure and silage. *Lactobacillus*species are used to produce cheese, yogurt, sour milks and are also found usefulin fermentation of vedgetables to produce pickles and sauerkraut, beverages such as wine and juices, some sausagesand sourdough breads (Osset et al., 2001; Aneja et al., 2014). During fermentation, these Lactobacillus species do also produce lactic acid as end product.(Osset et al., 2001; Miele et al., 2009). Lactic acid bacteria are more frequently found in unpasteurized juices (Oliveira et al., 2006). These bacteria species produces formic acid and acetic acid along with crborn dioxide and ethanol which can alter the flavour of juice (Jay and Anderson, 2001). *Aeromonas hydrophila* is a Gram negative bacteria that produces gas from fermented sugars while *Staphylococcus aureus* is a Gram positive bacteria that is normally associated with the human body. However, the use of plants' extracts was able to salvage the prepared juices from the havoc these microorganisms are known to bestow on juice for rejection and unacceptability. Different methods are used for the preservation of fruits and fruit products to inactivate enzymes that can degrade juice qualities and to also inhibit or eliminate spoilage microorganisms.

Table 3. Microbial load (CFU/ml); (spore/ml) in preserved fruit juice samples

	Bacteria	Fungi	Bacteria	Fungi	Bacteria	Fungi
Pasteurized juice before preservatives at day 0						
AJBP	66×10^4	54×10^3				
PJPB	99×10^4	63×10^3				
AJAP	25×10^4	20×10^3				
PJAP	42×10^4	39×10^3				
Pasteurized juice with preservatives from day 0						
	Day 0		After 5 days		After 10 days	
AJGAP	25×10^4	20×10^3	15×10^4	14×10^3	5×10^4	7×10^3
PJGAP	42×10^4	39×10^3	11×10^4	15×10^3	3×10^4	9×10^3
AJNLP	25×10^4	20×10^3	19×10^4	16×10^3	11×10^4	10×10^3
PJNLP	42×10^4	39×10^3	25×10^4	18×10^3	14×10^4	12×10^3
AJSBP	25×10^4	20×10^3	10×10^4	8×10^3	6×10^4	5×10^3
PJSBP	42×10^4	39×10^3	8×10^4	15×10^3	6×10^4	8×10^3
RAJC	25×10^4	20×10^3	10×10^4	10×10^3	18×10^4	15×10^3
RPJC	42×10^4	39×10^3	10×10^4	11×10^3	16×10^4	19×10^3

Legend**:** Apple juice before pasteurization (AJBP), Pineapple juice before pasteurization (PJBP), Apple juice *Gmelina arborea* preserved (AJGAP), Pineapple juice *Gmelin aarborea* preserved (PJGAP), Apple juice *Nauclea latifolia* preserved (AJNLP), Pineapple juice *Nauclea latifolia* preserved (PJNP), Apple juice sodium benzoate preserved (AJSBP), Pineapple juice sodium benzoate preserved (PJSBP), Refrigerated Apple juice control (RAJC), Refrigerated Pineapple juice control (RPJC).

Sensory evaluation of fruit juice

The same letter contained in each column signifies insignificant difference at ($p\leq 0.05$). Nevertheless, significant differences occurred in some parameters evaluated. The juice samples were endorsed for acceptability hence none of the samples rating fell below average for partial acceptability according to international standard rating. *G. arborea* preserved pineapple juice sampled on the overall acceptability was rated highest with a score of 8.62, followed by sodium benzoate preserved pineapple juice with a score of 7.90 and finally, *N. latifolia* and pineapple control with similar score of 7.60 (Table 4). *G. arborea* preserved apple juice samples on the overall acceptability was rated highest with a score of 7.64, followed by sodium benzoate preserved apple juice with 7.62, *N. latifolia* preserved apple juice with 7.60and finally apple juice control (no preservative) with 7.50.

The total titratable acidity observed in the juice samples is represented in Figure 1. The recorded total titratable acidity of apple juice preserved with *N. latifolia* was between 0.55 to 0.68% from day zero to day 14. That of pineapple juice preserved with *N. latifolia* was between 0.42 to 0.51% from day zero to day 14. The titratable acidity of apple juice preserved with *G. arborea* was between 0.50 to 0.56% and that of pineapple preserved with *G. arborea* was between 0.34 to 0.47% (Figure 1). The titratable acidity of apple juice preserved with sodium benzoate was in the

range of 0.46 - 0.53% from day zero to day 14 and that of pineapple preserved with sodium benzoate was between 0.31 to 0.44% from day zero to day 14. The titratable acidity value of apple juice without preservative ranged from 0.38 - 0.47% and pineapple juice without preservative from 0.28 - 0.43% at day 0 to 10 day of preservation. The obvious factorsinfluencing spoilage of fruit juices include amount of nutrient available, suitable preservation methods, redox potential, pH,microbial activities and availability of water for hydration of materials as highlighted in the research work of Vantarakis et al. (2011), Aneja et al. (2014). Among these factors, availability of water for hydration of materials and pHare the most influential determinants affecting fruit juice spoilage(Aneja et al., 2014) and these spoilage may include off-flavours, CO_2 production and changes in the colour, texture and appearance in juice (Lawlor et al., 2009; Sospedra et al., 2012). In the sensory evaluation, the higher rating of *G. arborea* fruit extract over *N. latifolia*stem barkin preservation could be as a result of the preservative effects that kept microbes from interfering with the juices taste, color and aroma.

Table 4. Sensory evaluation of juice samples

Samples	Colour	Taste	Aroma	Overall acceptability	
AJNLP	7.54^{b}	7.46^{b}	7.56^{b}	7.60^{b}	
AJGAP	7.80^{b}		7.63^{b}	7.67^{b}	7.64^{b}
AJSBP	7.55^{b}		7.65^{c}	7.67^{b}	7.62^{b}
AJC	7.27^{c}		7.45^{c}	7.68^{b}	7.50^{c}
PJNLP	7.28^{c}		8.25^{a}	7.81^{b}	7.60^{b}
PJGAP	8.84^{a}		8.26^{a}	8.78^{a}	8.62^{a}
PJSBP	7.55^{b}		8.26^{a}	7.78^{b}	7.90^{b}
PJC	7.26^{c}		7.65^{b}	7.82^{b}	7.60^{b}

Legend: Apple juice *Nauclealatifolia* preserved (AJNLP),Apple juice *Gmelinaarborea* preserved (AJGAP), Apple juice sodium benzoate preserved (AJSBP), Apple juice control (AJC) Pineapple juice *Nauclealatifolia* preserved (PJNLP), Pineapple juice *Gmelinaarborea* preserved (PJGAP), Pineapple juice sodium benzoate preserved (PJSBP), Pineapple juice control (PJC).
abc signifies that means with different letters in a same parameter are significantly different from each other ($p \leq 0.05$). Each value is a mean standard deviation of triplicate determination per sample

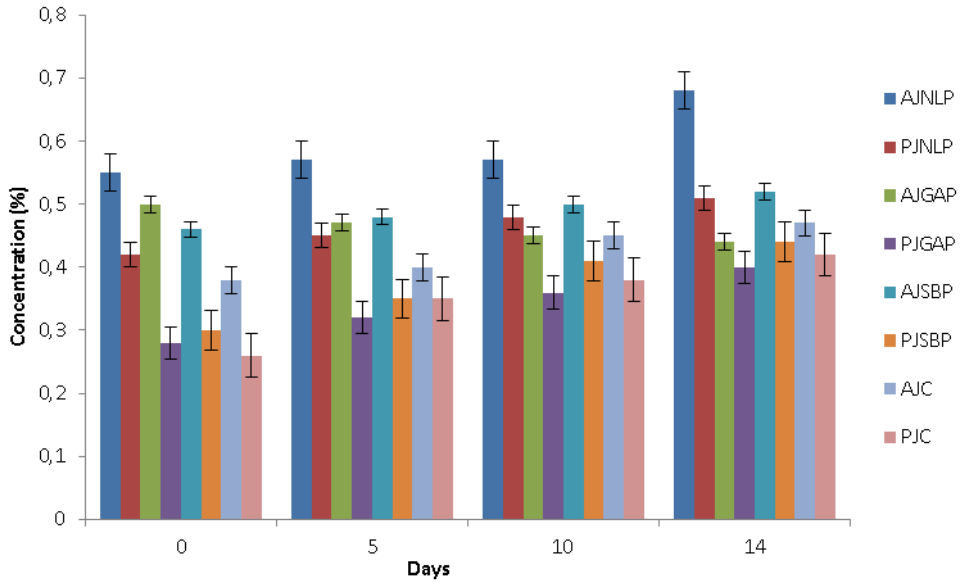

Figure 1.Titratable acidity of preserved juice.
Legend: Apple juice *Nauclealatifolia* preserved (AJNLP), Pineapple juice *Nauclea latifolia* preserved (PJNLP), Apple juice *Gmelinaarborea* preserved (AJGAP), Pineapple juice *Gmelina arborea* preserved (PJGAP), Apple juice sodium benzoate preserved (AJSBP), Pineapple juice sodium benzoate preserved (PJSBP), Apple juice control (AJC), Pineapple juice control (PJC).

In this study, *G. arborea* preserved pineapple and apple juices had the lowest acidity level with values of 0.40% and 0.41% respectively, while *N. latifolia* preserved juice had acidity level of 0.68% for apple juice and 0.51% for pineapple. This could be due to resident microorganisms fermenting the available sugar constituentsin the fruit juices which also reflected in the sensory evaluation for a reduced preservative quality. Decrease values of titratable acidity in the juice was as a result of reactions between chemical andorganic constituents contained in the fruit juicewhich was influencedcertainly by enzyme activities and storage temperature (Mehta and Bajaj, 1993; Parreek et al., 2011), lemon (Palaniswamy and Muthukrishnan, 1974) and aonla pulp and juice (Singh et al., 1998; Jain and Khurdiya, 2009). The low level of acidity in *G. arborea* preserved juice contributes to the flavour but it was in part responsible for the excellent stability against microorganisms.

CONCLUSIONS

Due to the obtained results, *G. arborea* is suggested to be a more potential preservative than *N. latifolia*. Previous studies on the use of *N. latifolia* and *G. arborea* as a preservative agent have not been documented and have demonstrated chances of being able to enhance shelf life of apple and pineapple juices. Major challenges in juice as fresh food are their limited storage life and their association with pathogens, resulting in continuing commercial pressures to use synthetic chemicals as preservatives. Natural remedies as found in plants are with little or no negative health consequences that can be exploited by food industries to overcome the incessant challenges poised on foods by microorganisms. The employment of plant antimicrobials to extend the storage of fruit juice will help to overcome spoilage and residual toxicity caused by synthesized chemical preservatives. This study on the antimicrobial of two candidate's natural preservative sources from *G. arborea* and *N. latifolia*, has helped to suggest the use of the plant extracts as potent plants to employed as natural preservatives on fruit juice as the obtained results can be compared with the preservative effect of a chemical preservative (sodium benzoate. This study, has suggested the extract of *G. arborea* as a promising preservative than that of *N. latifolia* in fruit juice preservation as suggested with the comparable results to that of sodium benzoate.

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