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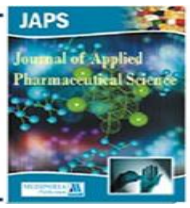

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The effect of ginger and garlic on the microbial load and shelf life of Kunun-zaki

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ABSTRACT

Kunun-zaki was produced using ginger and garlic and stored under ambient conditions for 10 days. The microbiological load and the shelf life of the drink were investigated. Diverse microbial genera: *Lactobacillus, Bacillus, Staphylococcus, Aspergillus, Penicillum, Fusarium and Saccharomyces* were isolated from samples. The effect of ginger and garlic separately were compared to the combined effect of ginger and garlic in reducing the microbial population. Of all the treatments, garlic (2g in 200mls or 0.01%$^{w}/_{v}$) was most effective in reducing the microbial populations. In contrast, treatment with 1g of ginger was least effective in reducing the microbial populations. Shelf life based on sensory overall acceptability and microbial quality of the samples varied with treatments but combination treatment with 2g in 200mls(0.01%$^{w}/_{v}$) ginger and garlic extended the shelf life by approximately four (4) days whereas other treated samples showed marginal enhanced shelf life of 2days. However, untreated control sample exhibited remarkably high microbial load and was virtually unacceptable after 24h of production. The result shows the potential of the combination treatment of ginger and garlic as antimicrobials and in extending the shelf life of Kunun-zaki.

Keywords: Kunun-zaki, ginger, garlic, shelf life, microbial load.

INTRODUCTION

Kunun-zaki, a non alcoholic beverage, very popular, especially in Nothern Nigeria is known for its social, religious and therapeutic values. It is taken as a refreshing drink, appetizer, food compliment, thirst quencher, substitute for or to complement soft drinks and wines at social gatherings. (Oramili *et al;* 2003). The nutritional value of Kunun-zaki as documented by Ayo and Okaka (1998) and Sopade and Kassun (1992) justifies its use. It is found to be rich in carbohydrate, vitamins and minerals and contains protein and fat. Sweet potato, which is used as an additive in its preparation, adds to the vitamin content. Kunun-zaki is produced from fermented millet, sorghum, guinea corn and maize in decreasing order of preference and is normally flavoured with ginger. It is consumed in the active state of fermentation.

The fermentation process is normally dominated by the microbial genera; *Lactobacillus, Bacillus, Aspergillus and Saccharomyces*. (Efiuvwevwere and Akoma, 1997). The method of preparation is simple and cheap and does not require any elaborate equipment. (Agboola, 1987) The short shelf life of Kunun-zaki is a major problem of both the brewers and consumers of this drink. These deleterious changes are due primarily to the objectionable off-flavour (over souring) induced by microorganisms. The use of chemical preservatives for microbial control in beverages is desirable and continues to generate research interest worldwide. However, with most of the published information being on the use of inorganic chemical preservatives to preserve this product, the objective of this study is therefore based on the use of organic chemical preservatives (ginger and garlic) to enhance the shelf-life stability of the product while maintaining the characteristic organoleptic attributes.

MATERIALS AND METHODS

Sample collection

Millet grains (*Pennisetum typhoides*), sugar, dried pepper, ginger and garlic were obtained from Umuahia main market, Abia State, Nigeria.

Production

Kunun-zaki was produced using the method described by Efiuvwevwere and Akoma (1995). Millet grains (1000g) were cleaned and steeped in water (1.5L) for 24h at ambient temperature. After 24h, the water was decanted off and the grains washed with tap water before blending with 10g of dried pepper in two volume tap water. The slurry was sieved and the filtrate allowed to sediment for 3-5h at ambient temperature. The supernatant was discarded while the various treatments were added to the remaining milky and pasty sediment. Each of the different concentrations of the treated samples and the control were divided into two portions. One of these was gelled using two volume boiling water and then allowed to cool to between 45^{o}C -50^{o}C. The gelled portions of each of the different concentrations of the samples and control were mixed (1:1 by volume) with the ungelled portion. Each of the mixture was then diluted with tap water, allowed to ferment for 8h and then sweetened with granulated sugar. The samples were dispensed into sterile screw capped containers.

Storage

The samples were stored at ambient temperature (25-32^{o}C) for 10 days.

Enumeration and isolation of microorganisms

This was carried out on Nutrient agar (NA), Sabouraud Dextrose agar (SDA), and Rogosa agar (oxoid) using pour plate method. The samples were serially diluted and 1ml of appropriate dilution was used to inoculate each of the agar plates in triplicates. The nutrient and rogosa agar plates were incubated at 37^{o}C aerobically for 24-36h while the SDA plates were incubated at room temperature for 48-72h. Colonies were counted using a colony counter (Gallenkamp). The mean of duplicate results were then recorded as the colony count. (Lateef *et al*., 2004).

Identification of isolates

Discrete colonies were picked at random, purified and characterized based on standard microbiological cultural, morphological and biochemical characteristics. (Harrigan and McCance, 1976 and Sneath *et al.*,1986). The biochemical tests include; catalase, oxidase, Voges-Proskauer, hydrogen sulphide production, nitrate reduction, citrate utilization and sugar fermentation.

For the fungi, the colonies were screened and identified based on the taxonomic schemes and descriptions by Fawole and Oso, (1998) and Mislivec *et al*., (1992)

Sensory analysis

The sensory attributes were evaluated as described by Larmond (1977).

Physicochemical analysis

pH and titrable acidity were determined. The pH was determined using referenced pH meter (model 291MK2, PYE UNICAM, England) while the titrable acidity was determined using the method described by Speck (1984)

RESULTS AND DISCUSSION

The rapid deterioration in shelf life of traditionally produced Kunun-zaki and other African beverages is widely acknowledged and is of great concern (Odunfa, 1985; Dirar, 1993; Efiuvwevwere and Akoma, 1995). The occurrence of diverse microbial genera and the remarkable high microbial load in the untreated control sample (Tables 1,2 and 3) are major cause of accelerated spoilage commonly experienced by brewers and consumers of these products. It is evident from tables 1,2 and 3 that the treated samples have lower microbial load because the added spices (ginger and garlic) have antimicrobial effect and are capable of destroying pathogenic bacteria (Ayo *et al.*, 2003).

In general, ginger and garlic have comparable antimicrobial activities due to the presence of essential oils in them (Nakatani, 2000, Ilondu *et al.*, 2001). But the more appreciable inhibitory effect exhibited by garlic (Tables 1,2, and 3) may be attributed to the differences in their essential oil components. Garlic tends to exert a more pronounced cell membrane interface and disruption than ginger due to the action of Allistatin I and Allistatin II (diallydisulphide oxide) contained in it which are not present in ginger and which affect the growth and respiration of microorganisms (Tynecka and Gos, 1973). Whereas the treatment with ginger and garlic separately resulted in reduction in the microbial load, the combination of the two (ginger and garlic) showed comparable efficacy when compared to that of treatment with ginger alone.

However, the magnitude of effectiveness of ginger and garlic as organic preservative on the microbial load of the sample

differed with the concentration. A decrease in the number of microorganisms was observed with the increase in the concentration of the preservatives. But the effects of ginger and garlic tended to decrease with storage time and this could be due to microbial degradation particularly in the presence of high lactobacilli load (De Boer, 1998).

Similar findings had been reported by Efiuvwevwere and Akoma, (1997) during Kunun-zaki preservation using inorganic preservatives. The drop in pH of all samples over the six days of storage (Table 4.) was in line with what was reported by Agarry *et al*.(2010). This may be as a result of greater microbial activities since this decrease was more observed in the untreated control samples having high microbial load. The high rate of change in pH with storage days could be due to the decomposition of fermentable substrates and sugars by microorganisms especially *Lactobacillus* species which ferment carbohydrates to produce energy and principally lactic acid.

The sensory analysis showed that the treated samples were more acceptable than the untreated control sample. The high rate of acceptability by the 10-member panel for the treated samples is attributed to the extra flavour added by the spices (Adeyemi and Umar, 1994). Combination treatment of ginger and garlic were more acceptable by the panelists with acceptability increasing with concentration and it extended the shelf life by about four (4) days. However, Kunun-zaki with garlic treatment alone showed apparent beneficial antimicrobial effects, but consumer acceptability was lower, confirming the off flavour that occurred when the concentration was increased.

The bacteria isolated included *Staphylococcus aureus, Lactobacillus* and *Bacillus* species. The dominance of *Lactobacillus* species in the samples may be due to the acidic nature of the drink and the presence of ginger is known to encourage its growth. Also, this could be as a result of one or more substances such as organic acids, ethanol, hydrogen peroxide and bacteriocins produced by *Lactobacillus* species during growth which inhibit or destroy the growth of other competitive microorganisms and thus improving the safety and keeping quality of such food products. This dominance made Efiuvwevwere and Akoma (1997) to conclude that Kunun-zaki is a lactic acid bacteria fermented product. The presence of *Bacillus* species in the samples is not accidental as they occur always on food of low acid content like drinks where they produce organic acids. They may also enter by soil contamination.

Consequently, the isolation of *Staphylococcus aureus* may be attributed to processing and post processing contamination through processors and utensils. Fungi isolated were *Aspergillus, Penicillium, Fusarium* and *Saccharomyces* species. The presence of these fungi is associated with spoilage of beverages. *Saccharomyces* spp are known to convert starch and dextrin to glucose which is then fermented to ethanol and carbondioxide.

Table. 1: Effect of ginger and garlic on the total viable count of Kunun-zaki during tropical ambient storage.

Storage (day)	K+GG 1g	K+GG 2g	K+GL 1g	K+GL 2g	K+GG+GL 0.5g each	K+GG+GL 1g each	Control
				10^4cfu/ml			
0	4.9	4.7	4.3	3.8	4.6	4.2	5.3
2	6.5	5.5	5.4	4.5	5.7	5.0	7.2
4	2.8	25	26	22	27	24	ND
6	30	28	28	25	26	27	ND
8	28	26	25	23	26	25	ND
10	23	21	19	16	20	21	ND

KEY:
K = Kunun-zaki, GG = Ginger, GL = Garlic, Control (contains neither ginger nor garlic), ND = Not Determined

Table. 2: Effect of ginger and garlic on the Total fungal counts of Kunun-zaki.

Storage (day)	K+GG 1g	K+GG 2g	K+GL 1g	K+GL 2g	K+GG+GL 0.5g each	K+GG+GL 1g each	Control
				10^4cfu/ml			
0	4.0	3.9	3.4	3.0	3.6	3.5	4.3
2	5.8	5.0	4.2	3.3	5.0	3.9	64
4	22	19	16	15	19	17	26
6	26	23	19	17	25	20	30
8	16	13	13	11	14	12	24
10	12	10	11	10	12	10	20

KEY:
KZ = Kunun-zaki, GG = Ginger, GL = Garlic, Control (contains neither ginger nor garlic), ND = Not Determined

Table. 3: Effect of ginger and garlic on the *Lactobacilli* count.

Storage (day)	K+GG 1g	K+GG 2g	K+GL 1g	K+GL 2g	K+GG+GL 0.5g each	K+GG+GL 1g each	Control
				10^4cfu/ml			
0	4.3	4.1	3.6	3.2	3.9	3.8	4.6
2	6.0	5.2	4.5	3.5	5.3	4.0	70
4	28	26	25	18	27	22	30
6	30	29	29	22	29	26	ND
8	27	25	23	20	25	23	ND
10	21	17	17	13	19	16	ND

KEY:
KZ = Kunun-zaki, GG = Ginger, GL = Garlic, Control (contains neither ginger nor garlic), ND = Not Determined

Table. 4: Effect of ginger and garlic on the pH of Kunun-zaki.

Storage (day)	K+GG 1g	K+GG 2g	K+GL 1g	K+GL 2g	K+GG+GL 0.5g each	K+GG+GL 1g each	Control
0	4.65	4.69	4.74	4.82	4.70	4.76	4.62
2	4.05	4.09	4.14	4.20	4.10	4.16	3.56
4	3.01	3.04	3.10	3.18	3.06	3.12	2.20
6	2.25	2.29	2.33	2.38	2.29	2.34	1.20
8	2.20	2.22	2.25	2.30	2.23	2.27	ND
10	2.10	2.14	2.18	2.20	2.14	2.16	ND

KEY:
KZ = Kunun-zaki, GG = Ginger, GL = Garlic, Control (contains neither ginger nor garlic), ND = Not Determined

Table. 5: Effect of ginger and garlic on overall sensory acceptability of Kunun-zaki.

Storage (day)	K+GG 1g	K+GG 2g	K+GL 1g	K+GL 2g	K+GG+GL 0.5g each	K+GG+GL 1g each	Control
0	2.44	2.48	2.50	2.46	2.30	2.10	2.25
2	3.20	3.10	3.00	2.84	2.68	2.60	4.62
4	5.80	5.48	5.26	4.40	3.25	3.06	6.50
6	6.08	6.00	5.89	5.70	5.52	5.40	7.00
8	7.22	7.08	6.88	6.40	6.15	6.10	ND
10	ND	ND	ND	7.50	7.20	7.02	ND

KEY:
KZ = Kunun-zaki, GG = Ginger, GL = Garlic, Control (contains neither ginger nor garlic), ND = Not Determined

CONCLUSION

From this study, the combined treatment of garlic and ginger, especially the 0.01(w/v) extended the shelf life of Kunun zaki up to about 4days. The shelf life extension means enhanced commercial potential of Kunun zaki. The combined treatment with ginger and garlic especially the 0.01 (w/v) is therefore recommended.

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Two hundred samples of freshly formulated Kunun-zaki, a locally fermented non-alcoholic cereal beverages were obtained from local hawkers in five (5) different locations in Chikun Local Government of Kaduna State, Nigeria and screened for enteric bacteria. They were analyzed using the Spread Plate Method. The pH of the samples ranged between 3.00 -7.50 and total bacterial count ranged between 2.0 Ã— 104 â€“ 9.0Ã—105 cfu/ml. The microorganisms recovered were Esherichia coli, Proteus vulgaris, Proteus mirabilis and Citrobacter freundii.Â Effect of chemical treatment and pasteurization on the shelf life of Kunun zaki (sorghum and maize gruel). European Journal of Food 1: 61-70. [13]. Microbial analysis Bacterial load: The bacterial loads for all samples were taken after. incubation for 18 hours. The bacteria loads were recorded for samples in both room temperature and low temperature for the period of 3 days (72 hours). The bacterial loads of samples are shown in the Table 6. The bacterial load of the Kunu sample containing lime and lemon was significantly ($P < 0.05$) higher than the bacterial load of all other samples on the first day at room temperature.Â 3. Adeyemi IA, Umar S (1994) Effect of method of manufacture on quality characteristics of kunun zaki, a millet based beverage. Niger Food J 12: 34-40. 4. Onuorah SI, Adesiyun AA, Adekeye JO (1987) Survival and multiplication of Staphylococcus aureus and Escherichia coli in Nigerian cereal drink (kunun zaki).