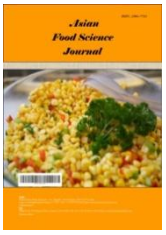

Asian Food Science Journal

20(7): 6-17, 2021; Article no.AFSJ.67459
ISSN: 2581-7752

Inherent Microorganisms Affects the Quality of a Nigerian Fermented Beverage *"Agadagidi"* During Production

K. J. Mogaji[1*], D. J. Arotupin[1], O. Mogaji [1], A. O. Arogunjo[1], O. B. AJayi-Moses[1], I. M. Akinwunmi[1], P. O. Gabriel[1], E. S. Orekoya[1], O. L. Olajesu[1], H. J. Adeleye[1] and J. O. Aribisala[1]

[1]Department of Microbiology, Federal University of Technology, Akure, Nigeria.

Authors' contributions

This work was carried out in collaboration among all authors. The protocol and first draft of the manuscript was written by authors KJM and DJA designed this research. Second draft and corrections were jointly performed by authors OM and AOA Literature searches were managed by author ESO, HJA and OLO. Authors POG and IMA managed the analyses of the study. Authors OBAM and JOA performed the statistical analyses. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/AFSJ/2021/v20i730316
Editor(s):
(1) Dr. Cesar Ozuna Lopez, University of Guanajuato, Mexico.
(2) Dr. Mindaugas Liaudanskas, Lithuanian University of Health Sciences, Lithuania.
(3) Dr. Nelson Pérez Guerra, University of Vigo, Spain.
Reviewers:
(1) Jitendra Upadhyaya, McGill University, Canada.
(2) Antonio Caporusso, University of Basilicata, Italy.
Complete Peer review History: http://www.sdiarticle4.com/review-history/67459

Original Research Article

Received 16 March 2021
Accepted 26 May 2021
Published 02 June 2021

ABSTRACT

This research produced wine from unripe plantain fruits using spontaneous fermentation method. The period of production was five days. The purpose of this study was to isolate and identify microorganisms associated with unripe plantain fruits, determine the physicochemical parameters of the fermenting medium. Microbial count, foaming density, alcohol content, enzymes assay, sensory analysis, physicochemical properties, mineral content, antioxidants properties, and antinutrient content of the sample were investigated every 24 h for 5 days. A total of seven bacteria and four fungi consisting of yeasts and moulds were isolated during the study. The microbial loads of bacteria and fungi of the unfermented plantain fruits were $8.0 \times 10^6 \pm 0.01$ cfu/mL and 14.2×10^4

**Corresponding author: Email: kjmogaji@gmail.com;*

± 0.01 sfu/mL respectively. The temperature (ºC), pH and titratable acidity (%) ranged from 25.55-32.50, 4.51-5.50 and 0.99-3.50 respectively. The sample was observed to be colourless all through the fermentation periods. Turbidity of the samples increased during fermentation from 43.50 to 111.00. The data obtained from this work has shown the importance of unripe plantain micro-flora in the production of "Agadagidi". It is also concluded that consortium of microorganisms inherent within the plantain fruits are involved in plantain fruits fermentation. This information can contribute to a better understanding of the "Agadagidi" production process for a consistent quality beverage.

Keywords: "Agadagidi"; local alcoholic beverages; ripe and unripe plantain; plantain wine; microbial loads; microflora.

1. INTRODUCTION

Plantain (*Musa paradisiaca* Linn) is one of the most important staple food crops for millions of people both in developed and developing countries. It is one of the foods commonly consumed in the West Africa sub-region, North America and the Caribbean [1]. In West and central Africa, more than 10 million tons are produced annually and are traded locally [2]. In Nigeria, its consumption cuts across the indigenous groups and the numerous socio-economic classes because of the ease of preparation and consumption [3]. ''Agadagidi,'' a cloudy effervescent sweet-sour taste typical African traditional alcoholic beverage is made from overripe bananas and plantains through fermentation. It is common in south-western part of Nigeria [4].

Fermentation is a metabolic process that converts sugar to acid and alcohol through the actions of molds, yeasts and bacteria [5]. Fermentation is an age long method of processing cereals and legumes. Food fermentation, especially lactic acid fermentation is an important technology in Africa [6]. The technology is indigenous and is adaptable to the culture of the people [7]. The fermentation process meets the requirements of being low-cost, preventing food spoilage and food-borne diseases with respect to consumers living in a climate that aids the rapid deterioration of food [8-9]. In addition, fermented foods are of particular importance in ensuring adequate intake of protein and/or calories in the diet [10-11]. Fermentation actually holds promise as a food processing method that can be used to diversify the food uses of some under exploited plant foods like plantain [12]. The traditional processing of these foods needs to be changed or modified to improve their nutritional status [13].

The techniques used in the production of wines from tropical fruits are similar to those of grape wine production which include pressing out the juice, fermenting, maturing and bottling [14]. Fermentation leads to changes in appearance of food which is characterized by different reactions of microorganisms [15]. Fermentation of foods covering a wide range of microbial and enzymatic processing of foods and ingredients is used to achieve desirable characteristics such as prolonged shelf-life, improved safety, attractive flavour, nutritional enrichment and promotion of health [16,17].

2. MATERIALS AND METHODS

2.1 Collection of Sample

Unripe plantain fruits (*Musa paradisiaca*, Linn) were obtained from a farmland in Arigidi-Akoko, Ondo State, Nigeria. The plantain fruits were kept in a sterile air tight polythene bags and transported to the Microbiology Postgraduate Laboratory, Federal University of Technology, Akure for further analysis.

2.2 Traditional Preparation of ''*Agadagidi''* Sample

The plantain fruits were divided into two portions; A and B. Portion A was left unfermented (control sample), while Portion B was fermented for 5 days.

The unripe plantain fruits were washed under running water to remove dirt, peeled and cut into small pieces. Sliced plantain of 28.6 grams were soaked in 500 mL of sterile water in a sterilized earthenware pot for five (5) days and were mashed. At the expiration of fermentation, the plantain fruit was then filtered through two layers of muslin cloth to remove the plantain pulp. The fermented alcoholic drink was then dispensed into sterile containers. The liquid then served as "Agadagidi".

The unfermented sample was withdrawn immediately after water was added into the plantain fruits.

2.3 Isolation and Identification of Microorganisms

Nutrient agar (NA), nutrient broth (NB), Potato dextrose agar (PDA), Potato dextrose broth (PDB) were prepared according to manufacturer's specification for the isolation of bacteria and fungi. At every 24 h for 5 days, samples were aseptically withdrawn and serially diluted for the isolation of bacteria and fungi. Microorganisms were incubated at 37°C and 27°C for bacteria and fungi respectively. Colonies and spore forming units formed on the media were counted and identified [18].

2.4 Determination of Colour

Colour determination was done using the method of "The Standard Lovibond Nessleriser Disc". Each sample of 5 mL was put in one tube of Lovibond Comparator and equal volume of distilled water was also put in the second tube. The colour was then read on the disc. The colours were expressed in Hazen Units (H.U) [19].

2.5 Determination of Total Titratable Acidity (TTA)

The total titratable acidity of the fermenting broth was determined every day as described by [20]. Exactly 10 mL of the sample was pipetted into a conical flask with 3 drops of phenolphthalein as an indicator was titrated against 0.1 M NaOH. The end point was noticed with the pink colour appearance due to phenolphthalein indicator. The volume of the end point was recorded when the pink colour was noted [21].

2.6 Determination of pH

The pH value of each sample were determined using Extech Instrument pH 100 after standardization with appropriate buffers (4 and 7 solutions). The electrode sensor of the pH meter was inserted directly into 20 mL sample in a clean 50 mL glass beaker. The electrode was then left in the media for 3 min to stabilize after which the pH were read.

2.7 Determination of Temperature

The temperature of fermenting sample was determined. The temperature was determined every 24 h using a portable temperature meter.

2.8 Determination of Turbidity

Turbidity was determined using the cell riser which was installed in the cell holder of model 2100A Turbidimeter in one of the two highest ranges (100 0r 1000 NTU). A clean sample cell of turbidimeter was filled with 25 mL of each sample and covered with light shield and turbidity read in Nephelometric Turbidity Units (NTU).

2.9 Statistical Analysis

The result of the experiment was in triplicate. The data obtained were subjected to one way analysis of variance (ANOVA) SPSS version 2.0. Differences were considered significant at $P<0.05$.

3. RESULTS

3.1 Isolation and Enumeration of Microorganisms Associated with Plantain Fruits

3.1.1 Total viable microbial counts from the samples

The total viable bacterial counts (10^6 cfu/mL) and fungal counts (10^4 sfu/mL) are shown in Figs 1 and 2 respectively. The total viable bacterial count of unfermented sample was 8.01 x 10^6 cfu/mL while that of fungi was 1.42 x 10^5 sfu/mL. Day 5 had the highest viable bacterial count 3.20 x 10^7 cfu/mL while Day 4 had the highest viable fungal count 3.00 x 10^5 sfu/mL.

3.1.2 Cultural, morphological, and biochemical characteristics of microorganisms isolated from the plantain fruits

The cultural, morphological, and biochemical characteristics of bacteria, yeasts, and moulds isolated from the plantain fruits are shown in Tables 1, 2 and 3 respectively. A total of 7 bacteria, 2 yeasts, and 2 moulds were isolated from the fermenting sample. The identified bacteria isolate from unripe plantain fruits during fermentation include; *Escherichia coli, Staphylococcus aureus, Bacillus subtilis, Bacillus cereus, Lactobacillus plantarum, Streptococcus feacalis* and *Leuconostoc mesenteriodes.*

The Moulds identified from unripe plantain fruits during fermentation include; *Aspergillus niger* and *Aspergillus flavus*. *Saccharomyces*

cerevisae and *Candida utilis* were the yeasts isolated during fermentation.

3.1.3 Microbial succession during fermentation

Table 4 shows the bacterial and fungal succession during fermentation of plantain unripe. *Bacillus* spp., *A. niger* and *A. flavus* were predominant at the beginning of the fermentation process. Day 2 to day 5 of the fermentation process, Lactic acid bacteria were the predominant bacteria involved in the fermentation while *Sacharomyces cerevisiae* and *Candida utilis* were the predominant fungi involved in the fermentation process.

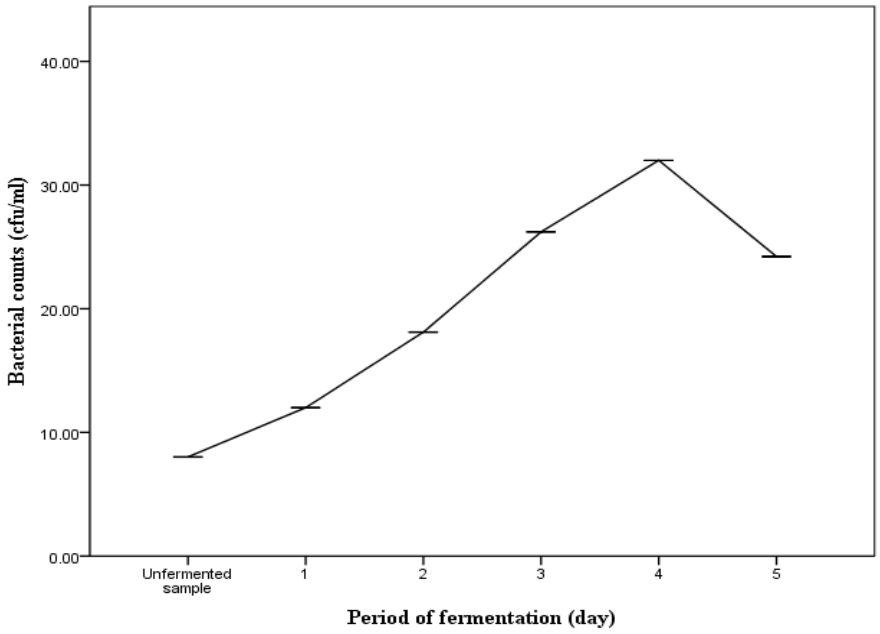

Fig. 1. Bacterial counts of unfermented and fermented unripe plantain samples

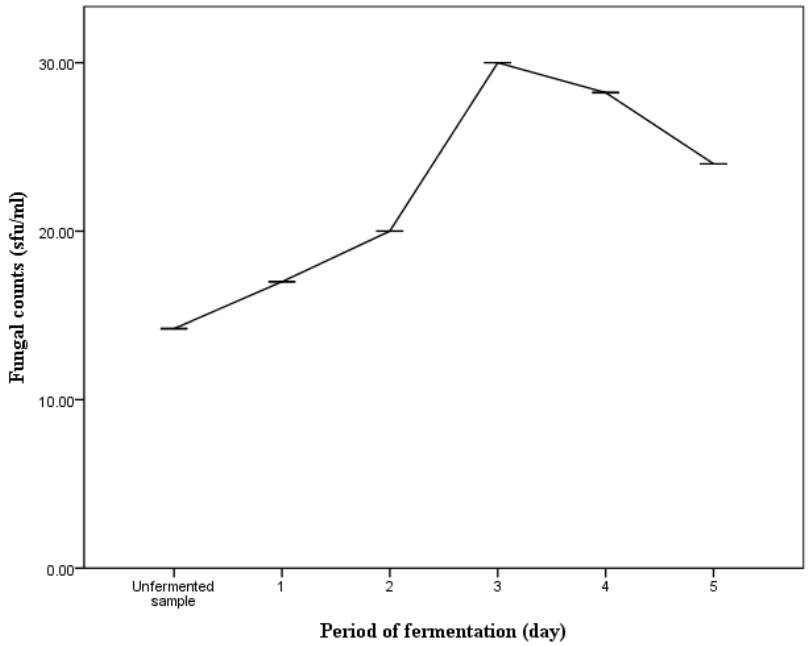

Fig. 2. Fungal counts of unfermented and fermented unripe plantain samples

Table 1. Morphological and biochemical characteristics of bacteria isolated from Samples

	A	B	C	D	E	F	G
Colour	Cream	Pale yellow	White	White	Yellow	Cream	White
Shape	Rod	Cocci	Rod	Rod	Cocci	Rod	Cocci
Edge	Entire	Entire	Irregular	Irregular	Entire	Irregular	Entire
Elevation	Flat	Raised	Flat	Flat	Raised	Circular	Circular
Surface	Rough	Smooth	Smooth	Rough	Smooth	Smooth	Smooth
Gram	-ve	+ve	+ve	+ve	+ve	+ve	+ve
Catalase	+ve	+ve	+ve	+ve	-ve	-ve	-ve
Coagulase	+ve	-ve	-ve	-ve	-ve	-ve	-ve
Spore	-ve	-ve	+ve	+ve	-ve	-ve	+ve
Motility	+ve	+ve	+ve	+ve	-ve	-ve	-ve
Oxidase	-ve	-ve	-ve	+ve	-ve	-ve	-ve
Indole	+ve	+ve	-ve	-ve	-ve	-ve	-ve
MethylRed	+ve	+ve	-ve	-ve	-ve	-ve	-ve
Urease	-ve	-ve	-ve	-ve	-ve	-ve	-ve
Starch	-ve	-ve	+ve	+ve	-ve	-ve	+ve
Glucose	AG	AG	A	A	-	AG	AG
Sucrose	-	A	A	A	-	-	AG
Maltose	A	A	A	A	-	A	AG
Lactose	AG	A	A	A	A	AG	AG
Fructose	-	A	AG	A	A	A	AG
Galactose	AG	AG	AG	-	-	AG	A

Key: A – Escherichia coli B - Staphylococcus aureus C – Bacilus subtilis
D – Bacilus cereus E - Streptococcus faecalis F–Lactobacilus Plantarum
G– Leuconostoc mesenteriodes, A= Acid present and gas absent, AG= Acid and gas present, -ve = Negative, +ve = Positive-=Absent

Table 2. Morphological and biochemical characteristics of yeast isolated from samples

	A	B
Cultural Characteristics		
Colour	Cream	Brown-black
Size	Medium	Medium
Surface	Smooth	Smooth
Shape	Oval	Cylindrical
Spore	+	+
Mycelium	-	-
Sugar Fermentation		
Glucose	FA	FA
Maltose	FA	FA
Lactose	A	-
Fructose	FA	FA
Sucrose	FA	FA
Galactose	FA	-

Key: A – *Saccharomyces cerevisiae* B - *Candida utilis*
A = Acid, FA = Fermentation and Assimilation, - = Absent, + = Presen

3.1.4 Physicochemical properties of samples during fermentation

There was no visible colour observed during fermentation periods.

Fig. 3 shows steady increment in temperature (°C) in both plantain fruit samples. The highest temperature value was on day 5 (32.50) of the fermentation period. Temperature value of unfermented sample was (25.54) which differed significantly (*P<0.05*) from the value obtained for day 1 to day 5 of fermentation. The highest pH value was obtained in unfermented sample (6.07), while the lowest pH was on day 5 (4.71).

The pH values of the unfermented sample and the fermented sample differed significantly (*P<0.05*) from each other (Fig. 4).

Fig. 5 shows that day 0 (unfermented sample) has the lowest titratable acidity (%) with a value of (0.99) while the highest was on day 5 with value of (3.50). There was significant difference (*P<0.05*) among the values of days; 1 to day 5 of the sample.

Variations in turbidity during fermentation of unripe and ripe plantain fruits are shown in Fig. 6. Sample had the turbidity ranging from 50.01±0.01 to 111.00±0.00 from day 1 to day 5. Turbidity gradually increased significantly (*p≤.0.05*) in the sample till the end of fermentation (Fig. 6).

4. DISCUSSION

The microbiological changes observed during fermentation could be as a result of diverse group of microorganisms present during fermentation process [22]. The bacterial count in the plantain fruit was higher compared to fungal count. This is in agreement with the report of Akinyosoye and Arotupin [23] stating that bacterial are most numerous organism in the soil. The high counts of bacteria, mould and yeast in the plantain fruits during fermentation may be due to the high concentration of nutrients which agrees with the findings of Arotupin et al. [24] that arrays of microorganisms could increase in population due to the nutrient rich nature of the substrate, thus, supporting the growth and proliferation of microorganisms. Low pH could be responsible for the increased fungal population [25]. The lignocellulosic structure of plantain fruits may be a factor responsible for the low microbial population in the unfermented sample [26].

The presence of bacteria such as *B. cereus,* and *B. subtilis* in the unfermented sample agrees with the findings of Oriola et al. [27]. The presence moulds such as *A. niger*, and *A. flavus* in the unfermented sample concurs with the finding of Oriola et al. [27] when *A. flavus, A. fumigatus* and *P. notatum* were isolated from uncrushed plantain fruits. The dominant population of *A. flavus* particularly in the unfermented sample may be due to the presence of nutrients available within the plantain fruits for utilization. Nasrin et al. [28] attested to this when combination of molasses and jackfruit were used as a substrate for mutant strain of *A. niger* for citric acid production. *Saccharomyces cerevisae* and *Aspergillus* spp. may as well be responsible for lactic acid production which concurs with the documentation of Ogbonnaya and Chukwu, [29] that some yeasts (*Saccharomyces*) and moulds (*Penicillium, Aspergillus* and *Botrytis*) produce lactic acid.

The decrease observed in bacterial load during plantain fruits fermentation on day 5 may be due to inhibitory effect of bioactive substances [30,31]. The higher prevalence of rod-shaped lactic acid bacteria in this study corroborated the study of Nwokoro and Chukwu [32] who reported that lactic acid bacteria commonly predominates during food fermentation. This is because they are aciduric in nature [32,33].

The appearance and increase in the yeast count after 24 h of fermentation is attributed to the decrease in the pH that creates conditions ideal for yeast growth. This is similar with the finding reported for other fermented beverages [34]. Roles of yeasts have also been reported to include improving the organoleptic qualities by producing different flavours and aroma in different foods [35]. The moulds (*Aspergillus flavus* and *Aspergillus niger*) isolated in the study are commonly present as contaminants in fruits and legumes and do not appear to play any significant important role in the fermentation [36,37].

The subsequent disappearance of moulds after 24 h observed in this study as well as previous studies [37,32,33] was probably due to the low oxygen tension in the fermenting matrix. It could also be due to the presence of organic acids especially lactic acid as it could be seen that the growth of lactic bacteria and yeasts increased gradually throughout fermentation while the numbers of moulds decreased. The growth of lactic acid bacteria and yeasts can inhibit the growth of moulds [38].

The *Bacillus* species isolated in this study which was *Bacillus cereus* and *Bacilus subtilis*. Previous reports [36,32] have also isolated *Bacillus* species in the fermentation and they have been reported to show saccharolytic activities [32]. These microorganisms persisted towards the end of the fermentation indicating that they continued the saccharification of starch to release sugars. The presence of lactic acid bacteria as dominant organisms may be due to their aciduric nature [39].

Table 3. Morphology characteristics of mould isolated from samples

Isolates	Cultural characteristics	Spores/conidia arrangement under the microscope	Identity of isolates
A1	The surface is rough. Spores are granular flat, often with radial grooves yellow at first but quickly become bright yellow green with age. Sizes of colonies are medium.	Conidia are globose to sub globose with pale green colour. Septate hyphae with long condiophore bearing the conidia	*Aspergillus flavus*
A2	The surface is smooth with brown mycelia growth. With whitish colonies large in size.	An upright conidiopore that terminates in a swelling, bearing phialides at the apex radiating from the entire surface. Conidial are one celled and densely packed. Spores are black.	*Aspergillus niger*

Table 4. Microbial succession during fermentation of plantain fruits

	Unfermented sample	Day 1	Day 2	Day 3	Day 4	Day 5
A	*Bacillus cereus, Bacillus subtilis, Escherichia coli*	*Staphylococcus aureus, Streptococcus feacalis*	*Staphylococcus aureus, Lactobacillus plantarum, Leuconostoc mesenteriodes*	*Lactobacillus plantarum, Bacillus subtilis. Streptococcus feacalis*	*Lactobacillus plantarum, Streptococcus faecalis*	*Lactobacillus plantarum*
B	*Aspergillus niger, Aspergillus flavus*	*Aspergillus flavus, Saccharomyces cerevisiae*	*Saccharomyces cerevisiae, Candida utilis*	*Candida utilis, Aspergillus niger Saccharomyces cerevisiae*	*Saccharomyces cerevisiae, Candida utilis*	*Saccharomyces cerevisiae, Candida utilis*

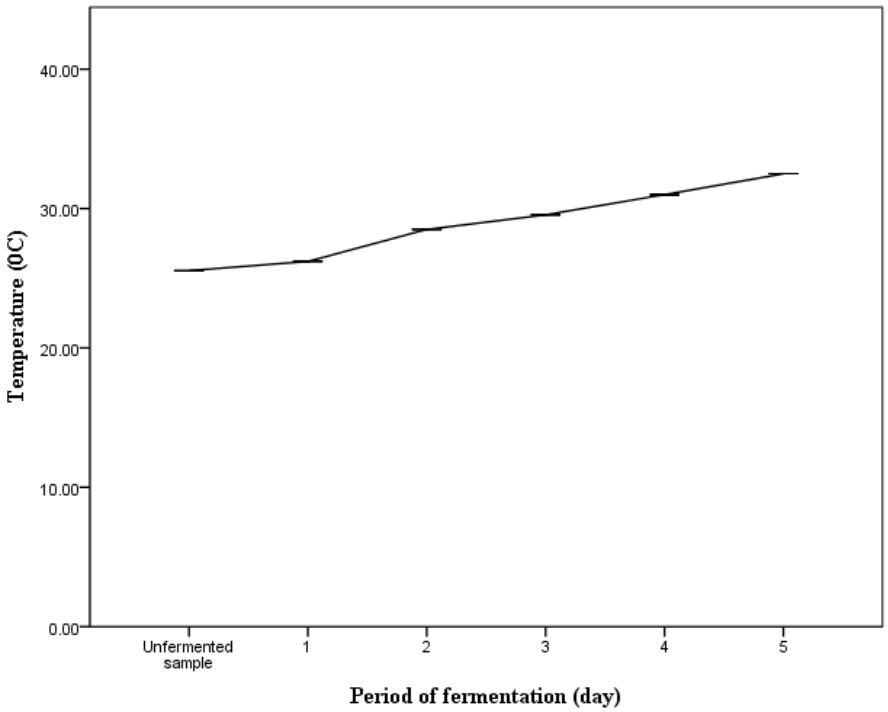

Fig. 3. Temperature of unfermented and fermented unripe plantain samples

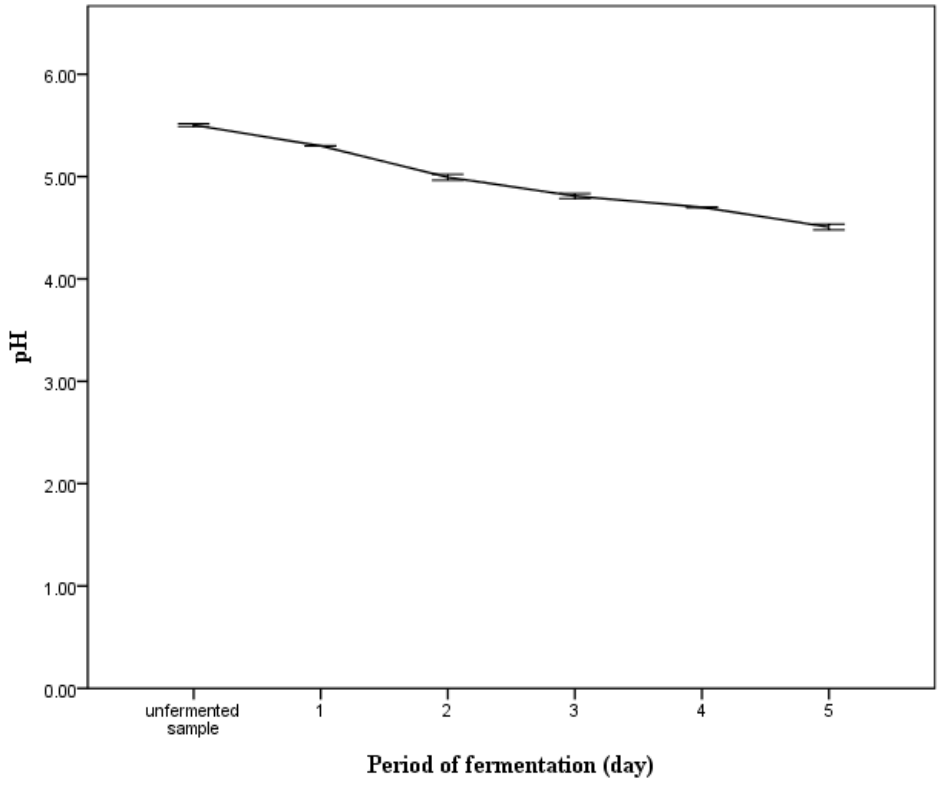

Fig. 4. pH of unfermented and fermented unripe plantain samples

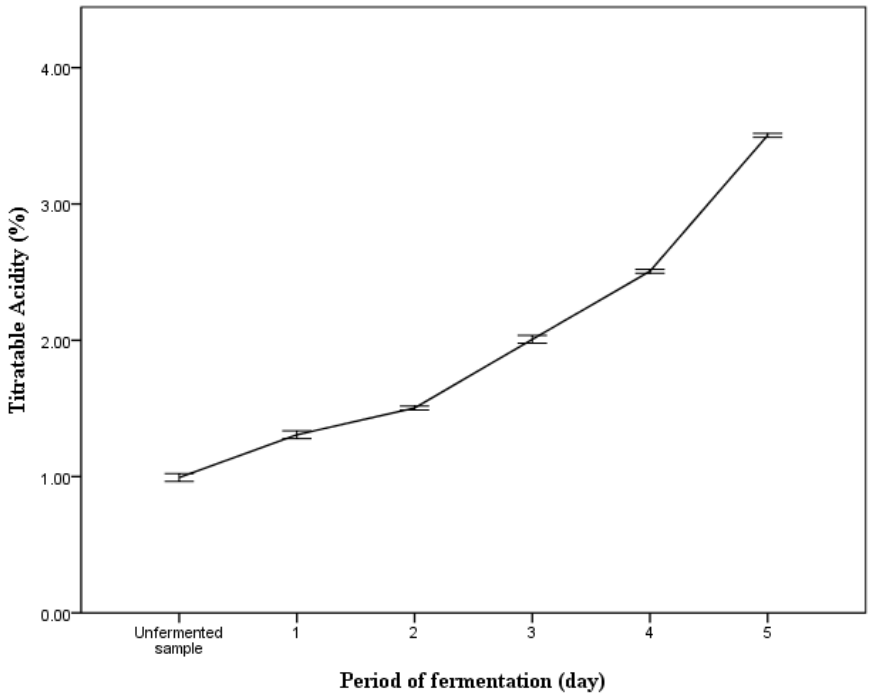

Fig. 5. Titratable acidity of unfermented and fermented unripe plantain samples

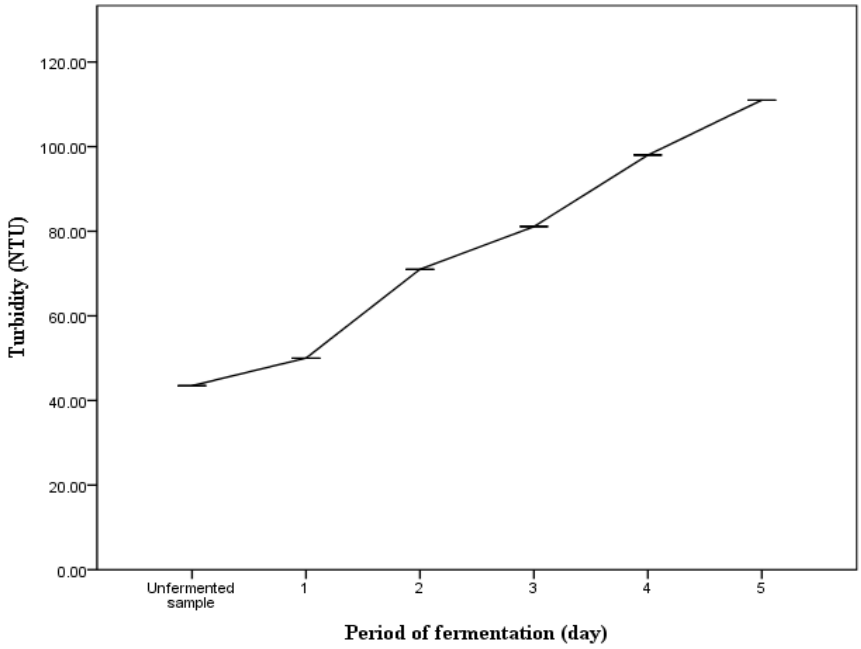

Fig. 6. Turbidity of unfermented and fermented unripe plantain samples

This study revealed that unfermented and fermented unripe plantain fruits colour was observed to be colourless. This could be as a result of the environment and natural colour of the fruits. There was a steady decrease in the pH during fermentation. This might be as a result of production of lactic acid by fermentative organisms (mainly lactic acid bacteria) responsible for the fermentation of "Agadagidi" [40]. A significant increase in the total titratable acidity (TTA) during fermentation was recorded in this study, and this could be as a result of

production of lactic acid and other organic acids by organisms responsible for the fermentation, as is the case with the decrease in pH. This observation of decrease in pH and increase in total titratable acidity (TTA) agrees with most fermentation studies including those of Wakil and Kazeem [41], Okwute and Olafiaji [42], and Modu et al. [43]. The early rise in titratable acidity and reduced pH is important to avoid proliferation of undesirable organisms resulting in poor fermentation [37]. During the process of fermentation, the temperature of samples was observed to increase as fermentation time progresses. The increases in temperature may be due to the activity of different microorganisms during fermentation process. This is similar to a result reported by [22], when he fermented ripe and unripe plantain flour. This supports the fact that fermentation is an exothermic process and that the heat generated was due to metabolic activities of microorganisms.

5. CONCLUSION

This study therefore showed that microorganisms such as *Escherichia coli, Staphylococcus aureus, Bacillus subtilis, Bacillus cereus, Lactobacillus plantarum, Streptococcus feacalis* and *Leuconostoc mesenteriodes, Aspergillus niger* and *Aspergillus flavus*, *Saccharomyces cerevisae* and *Candida utilis* were associated with plantain fruits. Thus, *Leuconostoc mesenteriodes, Lactobacillus plantarum, Bacillus subtilis, Saccharomyces cerevisae* and *Candida utilis* were found predominant in the production of plantain wine.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Padam BS, Tin HS, Chye FY, Abdullah MI. Banana by-products: an under-utilized renewable food biomass with great potential. Journal of food science and technology. 2014;51(12):3527–3545.
2. Nse-Nelson FA, Oke UR, Adindu JU. Analysis of plantain marketing in Ikwuano local government area of Abia State, Nigeria. Nigerian Journal of Agriculture, Food and Environment. 2016;12(1):85-89.
3. Solanke AO, Falade KO. A review of the uses and methods of processing banana and plantain (*Musa* spp.) into storable food products. Journal of Agricultural Research and Development. 2010;9(2):87-96.
4. Omojasola PF, Davies OF, Kayode RM. The effect of chemical preservatives, pasteurization and refrigeration on the shelf life of “Agadagidi” a fermented plantain drink. Research Journal of Microbiology. 2012;(3):145-157.
5. Anal AK. Quality ingredients and safety concerns for traditional fermented foods and beverages from Asia: A review *Fermentation*. 2019;5(1):8.
6. Siegel A, Faweett B. Food legume processing and utilization. International Development Research Centre Technical Studies Series, Ottawa; 1978.
7. Johansen PG, Owusu-Kwarteng J, Parkouda C, Padonou SW, Jespersen L. Occurrence and importance of yeasts in indigenous fermented food and beverages produced in Sub-Saharan Africa. Frontiers in microbiology. 2019;10:1789. Available:https://doi.org/10.3389/fmicb.2019.01789
8. Lorenzo JM, Munekata PE, Dominguez R, Pateiro M, Saraiva JA, Franco D. Main groups of microorganisms of relevance for food safety and stability: General aspects and overall description. Innovative Technologies for Food Preservation. 2018; 53–107.
9. Tortora Gerard J, Funke, Berdell R, Case Christine L. "5". Microbiology An Introduction (10 ed.). San Francisco, CA: Pearson Benjamin Cummings. 2010;135
10. Melini F, Melini V, Luziatelli F, Ficca AG, Ruzzi M. Health-Promoting Components in Fermented Foods: An Up-to-Date Systematic Review. Nutrients. 2018;11(5):1189.
11. Borresen EC, Henderson AJ, Kumar A, Weir TL, Ryan EP. Fermented foods: patented approaches and formulations for nutritional supplementation and health promotion. Recent patents on food, nutrition & agriculture. 2012;4(2):134–140.
12. Onweluzo JC, Nwabugwu CC. Fermentation of Millet (*Pennisetum americanum*) and Pigeon Pea (*Cajanus cajan*) Seeds for Flour Production: Effects on Composition and Selected Functional Properties. Pakistan Journal of Nutrition. 2009;8:737-744.
13. Mugendi JB, Njagi ENM, Kuria EN, Mwasaru MA, Mureithi JG, Apostolides Z. Effects of processing techniques on the

nutritional composition and anti-nutrient content of mucuna bean (*Mucuna pruriens* L.). African Journal of Science. 2014;(4): 156-166.

14. Okafor N. Modern industrial microbiology and biotechnology. Science Publishers. Enfield N. Y, U. S. A. 2007;530.
15. Ray M, Kuntal GS, Singh K, Chandra M. Folk to functional: An explorative overview of rice-based fermented foods and beverages in India. *Journal of Ethnic Foods*. 2016;23:5-18.
16. Tamang JP. Health benefits of fermented foods and beverages. CRC Press, Taylor and Francis Group, New York. 2015;1-110.
17. Areola JK, Yussuf IO. The processing and nutritional analysis of plantain wine. International Journal of Business and Educational Policies. 2011;7(2):238-245.
18. Fawole MO, Oso BA. Characterization of Bacteria: Laboratory Manual in Micro-biology. 5th Edition. New Spectrum Books Publisher, Ibadan, Nigeria. 2007;24-33.
19. APHA. Standard methods for the examination of water and waste water. 18thEdition. American Public Health Association, Washington. 2002;135-170.
20. AOAC. Official methods of analysis of the Association of Official Analytical Chemists international. 19th edition. Gathersburg, Maryland, U.S.A; 2012.
21. Mbajiuka CS, Obeagu EI, Chude CN, Ihezie OE. (2014). Antimirobial effects of *Chromolaena odorata* on some human pathogens International Journal of Current Microbiology and Applied Science. 2014;3(3):1006-1012.
22. Ojokoh Eromosele AO, Ojokoh O, Adewale Ekundayo Linda Ezem A, Anyiam Chukwudum. Effect of Fermentation on the Proximate Composition of Ripe and Unripe Plantain Flour. Journal of Advances in Microbiology. 2017;2(3):1-10, 2017.
Article no.JAMB.31303.
23. Arotupin DJ, Akinyosoye FA. Microbiologi-cal and physicochemical characteristics of cassava cultivated soils. Research Journal of Microbiology. 2008;3:41-46.
24. Arotupin DJ, Awojobi KO, Owolabi TR, Eze C. Microorganisms Associated with African Star Apple (*Chrysophylum albidum*) and their Hydrolases. African Journal of Microbiology Research. 2016;10(7):225-230.
25. Bello BK, Akinyele BJ. Effect of Fermentation on the Microbiology and Mineral Composition of an Edible Mushroom *Termitomyces robustus* (Fries). International Journal of Biological Chemistry. 2007;4:237-243.
26. Agbor VB, Cicek N, Sparling R. Biomass Pretreatment: Fundamentals toward Application. Biotechnology Advances. 2011;29:675–685.
27. Oriola O, Boboye B, Adetuyi F. Microorganisms associated with the production of a Nigerian Fermented Beverage, 'Agadagidi'. Microbiology Research Journal International. 2017;20(6): 1-9, Article no. MRJI.32654ISSN: 2456-7043.
28. Nasrin S, Ansary MU, Alam KM. Optimization of Citric Acid Production by Substrate Selection using Gamma Ray Induced Mutant Strain of Aspergillus niger. Global Journal of Medical Research: C Microbiology and Pathology. 2017;17(2): 22-30.
29. Ogbonnaya N, Chukwu BC. Studies on "Akamu", a traditional fermented maize food. Review on Children Nutrition. 2012;39:180-184.
30. Ouoba LII, Rechinger KB, Barkholt V, Diawara B, Traore AS, Jakobsen M. Degradation of proteins during the fermentation of African locust bean (*Parkia biglobosa*) by strains of *Bacillus subtilis* and *Bacillus pumilus* for production of Soumbala. Journal of Applied Microbiology. 2003;94:396-402.
31. Kalui CM, Mathara JM, Kutima PM. Probiotic potential of spontaneously fermented cereal based foods: A review. African Journal of Biotechnology. 2010;9:2490-2498.
32. Nwokoro O, Chukwu BC. Studies on 'Akamu', a tradditional fermented maize food. Revista Chilena de Nutrición. 2012;39(4):180–184.
33. Nsofor CA, Ume SC, Uzor BC. Isolation and characterization of lactic acid bacteria from 'ogi' sold in Elele, Nigeria. Journal of Biological and Food Science Research. 2014;3(2):19–22.
34. Abegaz KE, Beyene F, Langsrud T, Narvhus JA. Indigenous processing methods and raw materials of borde, an Ethiopian traditional fermented beverage. Journal of Food Technology in Africa. 2002;7(2):59–64.
35. Inyang CU, Idoko CA. Assessment of the quality of 'ogi' made from malted millet.

African Journal of Biotechnology. 2006;5(22):2334–2337.
36. Ogbulie JN. Production of Tempeh from African yam bean (*Sphenostylis stenocarpa* Harms). M. Sc Thesis, University of Port Harcourt, Nigeria. 1991;33.
37. Mbata TI, Ikenebomeh MJ, Alaneme JC. Studies on the microbiological, nutrient composition and antinutritional contents of fermented maize flour fortified with bambara groundnut (*Vigna sub- terranean* L.). African Journal of Food Science. 2009;3(6):165–171.
38. Edema MO, Sanni AI. Functional properties of selected starter cultures for sour maize bread. Food Microbiology. 2008:25: 616–625.
39. Kuta FA, Nimzing L, Orka'a PY. Screening of *Bacillus* species with potentials of antibiotics production. Food Biotechnology. 2009;24:42-46.
40. Oluwafemi F, Adetunji AF. Antimicrobial activities of lactic acid bacteria isolated from traditionally fermented maize ('ogi') against *Candida albicans*. Journal of Applied Biosciences. 2011;41:2820–2835.
41. Wakil SM, Kazeem MO. Quality assessment of weaning food produced from fermented cereal-legume blends using starters. International Food Research Journal. 2012;19(4):1679–1685.
42. Okwute LO, Olafiaji B. The effects of ginger (*Zingiber officinale*) on the microbial load of a Nigerian traditionally fermented maize paste ('ogi'). American Journal of Research Communication. 2013;1(9):84–98.
43. Modu S, Ibrahim Z, Hajjagana L, Falmata AS, Babagana M, Bintu BP. Production and evaluation of weaning meal from fermented red maize fortified with cowpea. Academia Journal of Food Research. 2013;1(3):050–058.

Peer-review history:
The peer review history for this paper can be accessed here:
http://www.sdiarticle4.com/review-history/67459