

Microbial, proximate and sensory quality of pito beverage locally prepared and hawked in Ogun State, Nigeria

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Abstract

Background and Objective: Pito is a traditionally brewed alcoholic beverage. It's processing is prone to microbial contamination. This study evaluated the microbiological, nutrient and sensory qualities of a locally hawked and laboratory prepared alcoholic beverage (Pito).

Materials and Methods: Samples were collected from four (4) different locations in Ogun State, and laboratory samples prepared as control were analyzed for proximate, microbiological and sensory properties using standard methods.

Result: The results of the proximate analysis show that the moisture, ash, fat, protein and carbohydrate content of the Pito beverage samples are significantly different with values that ranged from 90.12 to 95.42%, 0.74 to 0.93%, 0.16 to 0.32%, 0.41 to 0.58% and 3.21 to 7.58%, respectively. The protein, ash and fibre content of laboratory sample had higher values than the hawked samples. The results of the microbiological analysis shows that the Total viable count ranges from 4.1 to 6.3 x 10^6 cfu/ml, Coliform count ranges from 1.3 to 3.4 x 10^6 cfu/ml, *Salmonella* Count ranges from 1.3 to 2.4 x 10^6 cfu/ml, *Staphylococcus* Count were not detected in hawked freshly prepared Pito beverages obtained from Ifo and Agbara. Yeast Count ranges from 2.2 to 3.4 x 10^6 cfu/ml and Mould Count ranges from 1.2 to 3.3 x 10^6 cfu/ml. The microbes isolated were: *Streptococcus sp, Staphylococcus sp, Klebsiella sp, Micrococcus sp and Salmonella sp. and the fungi* were *Saccharomyces cerevisiae, Aspergillus sp, Penicillium sp, Sporotrichum sp, Rhizopus sp and Mucor sp.* Sensory evaluation shows that laboratory sample was more acceptable in all attributes rated.

Conclusion: This study showed that the beverages had higher microbial loads than the specifications set for beverages. Presence of pathogenic species in some of these beverages is of paramount concern. Therefore, periodic screening of hawked Pito and their brewers, coupled with regular training and health education of handlers in all aspects of food hygiene and safety is encouraged

Keywords: pito; beverage, hawked, bacteria, microbial, sensory

Introduction

Food is an essential ingredient for sustenance of life either of plants or animals. Its demand can therefore, not be overemphasized. In Countries like Nigeria, people depend mostly on indigenous technology for food preparations especially food of plant origin. Some of the food that originate from plant include beverages, notably, Zobo, Kunun, burukutu, palm wine and Pito drink. Pito is a nutritious traditional beverage mostly consumed by inhabitants of Nigeria. Currently, it is widely consumed across the country due to urbanization and migration. It is also consumed in certain parts of Northern Ghana (Ezekiel et al., 2014) ^[12] and other African countries but under different names. According to Ellis et al., (2005) [10], Pito can be preserved and stored for eight weeks if all the right conditions of preparation, storage and preservation are achieved. Pito is commonly brewed and consumed by people in the rural community using traditional methods. It is brownish dark in colour (Avicor et al., 2015)^[4].

Pito is produced mainly from the grains of guinea corn (*Sorghum vulgare and Sorghum bicolor*). Sorghum is one of the cereals cultivated in the tropical regions of Africa and is about the largest cultivated crop in the Northern Guinea Savanna areas of Nigeria

Sorghum is a gluten-free cereal grown in many African countries primarily as food crop with less than 5% of the

annual production commercially processed by the industry (Okoli, *et al.*, 2010)). Sorghum is a large variable genus with many cultivars (Kolawole *et al.*, 2007) ^[16]. It constitutes a major source of energy and protein for people in Asia and Africa and it serves as staple food for many of the world's poorest and least privileged people

Pito contains remarkable levels of proteins according to Taofeek et al., (2014) and therefore has potential to complement protein-based meals, thus its consumption is encouraged owing to its properties as a functional food. This was supported by Pang et al., (2012) when they reported the presence of active components of functional foods such as polyphenols, micronutrients, and macronutrients which possess physiological benefits beyond basic nutritional requirements in the drink and plays an important role in the prevention of diseases relating to metabolic imbalances such as gastrointestinal disorders, inflammation, obesity, hypertension, type 2 diabetes, obesity and even cancer (Pang et al., 2012). Kolawole et al. (2007) ^[16] reported the presence of essential minerals that included zinc (Zn), calcium (Ca), magnesium (Mg), and iron (Fe) even though in limited amounts, in the Pito beverage Holzapfel, (2002) ^[14]. This was confirmed in a similar report by Duod *uet al.*, (2012) ^[6] with the exception of manganese (Mn). The difference may be due to the variations in the source of the raw material. Minerals are very important for the up-keep of

the body tissues and helps in preventing depression. These health benefits may also be derived from sorghum being a major source of antioxidant and phytochemical constituents (Dykes et al., 2009)^[8]. In Nigeria, many women have been able to set up small scale commercial production of Pito which has helped to alleviate poverty among the people. However, the production of Pito for commercial purposes, it's processing is highly prone to microbial contamination. A large number of lactic acid bacteria, coliforms, molds and veast have been reportedly implicated in food spoilage as they use the carbohydrate content for fermentation processes (Amusa et al., 2005)^[2]. In developing nation like Nigeria, it has not been possible to have control over the processing of hawked foods, because most of the vendors lack the adequate knowledge of food processing and handling practices, there is a high risk of chemical and microbial contamination. In most Nigeria cities, the sales and consumption of this locally made beverage is high due to the high cost of other non-alcoholic drinks. However, this drink is usually hawked in the motor parks, school premises and market places. This research was conducted to investigate the microbiological and nutritional qualities of this hawked non-alcoholic drink called Pito in South Western Nigeria.

Materials and Method

Collections of Samples and Source of Raw Material

Hawked Pito drink samples were purchased from four locations in Ogun state namely, Lafenwa in Abeokuta,Sona in Sango, Abekoko in Ifo and Lusada market in Agbara all in South Western Nigeria. Sterile sample bottles were used, then properly labelled and were analysed immediately in the laboratory. Red colour sorghum was obtained from Sayedero market in Ilaro, Ogun state. The equipment to be used in producing pito was supplied by the Food Science and Technology Department, The Federal Polytechnic Ilaro, Ogun state.

Methodology

Preparation of Pito

The traditional method of brewing Pito goes through these basic stages. Malt Pito is made from sorghum. As describe by (Ellis et al., 2005) [10]. It involves soaking thirty kilogram of sorghum in water for 2days. The steeping liquor was chang e every 12hours to prevent odour. The grains are spread and allowed to germinate for 4days in red basket. On the 4th day the malt were packed into a nylon sack. and allowed to ferment. The geen malt were then sun dry and milled. The milled malt was mixed with about 1.5 llitres of water followed by a litrre of slimmy solution ymeswa the clear supernantant, which contain emzymes was decanted and coarse particle boiled 90oC for 30mins to gelatinize. The superntnantt was then mixed with the boiled coarse particle to attain temperatue of 60 °C. The mixture was then allow to undergo Lacic acid fermentation for 17hours and then filter to remove the coarse particle. The wort obtain

was boiled to evaporate excess water. The wort was cooled and pitched with yeast and allow to ferment for 15 hours. The product pito was cooled to 15° C to agglutinate the yeast and filter the resulting product is: a top clear supernatant called "Pito"

Proximate Analysis of Pito Samples

The moisture ash, fat, crude fibre and protein content of each sample was determined by the method described by the (AOAC, 2005)^[3].

The carbohydrate content was calculated by deducting the sum of the values for moisture, crude protein, crude fat, crude fibre and Ash in 100 (AOAC, 2005)^[3].

Microbial Analysis of Pito Samples

Isolation and identification of microbes: Ten (10) ml of each Pito samples were diluted in 90 ml of sterile distilled water in a conical flask to get the aliquot, a ten-fold serial dilution was carried out. An aliquot of 1 ml from selected dilutions of each sample was inoculated aseptically into labelled triplicate plates. The media used were (NA, for total viable count, MAC for total coliform, BPA for *Staphylococcus* count, BSA for *Salmonella* count) using standard pour plate method and incubated at 37°C ± 2 °C for 24 to 48 hours.

Potato Dextrose Agar was incubated at (28°C±2°C) for 3 to 5 days for isolation of fungi.

Colonies were enumerated at the end of incubation period using digital colony counter (Gallenkamp England). Microbial colonies were counted and recorded. Isolates were preserved on appropriate agar slants stored at 4°C for further analysis. Presumptive isolates were further characterized and identified on the bases of colonial morphology, microscopic and biochemical characteristics (Cheesbrough M., (2002) ^[5], Lynne, 2003) ^[17].

Sensory Evaluation of Pito Samples

The sensory evaluation of the Pito samples was carried out for consumer acceptance and preference using 10 semitrained panelist (students of Food Technology Department, The Federal Polytechnic, Ilaro). They were to evaluate the sensory properties based on Taste, flavor, crispiness, Appearance and Overall acceptability using a nine point Hedonic scale where 1 represents "extremely dislike" and 9 "extremely like" respectively.

Statistical analysis

All data obtained were subjected to statistical analysis of variance (ANOVA) using SPSS version statistical packages. Means were separated using DUNCAN Multiple Range Tests (DMRT).

Result and Discussion

Results

The microbial, proximate and sensory analyses of Pito are presented in Tables 1-3.

 Table 1: Proximate composition of Pito beverage locally prepared and hawked in five different locations in Ogun state, South Western Nigeria

Parameters	Samples					
%	Control	Abeokuta Sango		Ifo Agbara		
Moisture	95.42±0.05 ^a	93.22±0.07 ^a	90.83±0.04 ^a	90.12±0.03 ^a	92.42±0.01 ^a	
Ash	0.93±0.08 ^a	0.83±0.05 ^b	0.88±0.01 ^b	0.91±0.01 ^b	0.74±0.01°	
Fibre	0.00	0.00	0.00	0.00	0.00	

Fat	0.32±0.06 ^a	0.16±0.04°	0.25±0.01 ^b	0.22±0.01 ^b	0.23±0.02 ^b
Protein	0.58 ± 0.05^{a}	0.53±0.03ª	0.46 ± 0.09^{b}	0.44 ± 0.03^{b}	0.41±0.03 ^b
Carbohydrate	2.75±0.01°	5.26±0.03 ^b	7.58±0.08 ^a	8.31±0.01 ^a	6.20±0.06 ^b

Values are means + standard deviations of triplicate determinations. Means with the same superscript with a column are significantly different ($p \le 0.05$).

 Table 2: Microbiological analysis of Pito beverage locally prepared and hawked in five different locations Ogun state South Western Nigeria.

Parameters (x10 ⁶ cfu/ml)	Samples				
	Control	Abeokuta	Sango	Ifo	Agbara
Total viable Count	5.7±0.03a	6.3±0.03a	4.2±0.01b	4.1±0.05b	5.2±0.03a
Coli form Count	ND	3.4±0.08a2.	6±0.01b	2.0±0.02b	1.3±0.01c
Salmonella Count	ND	2.1±0.02b	2.4±0.01b	1.3±0.01c	2.1±0.03b
Staphylococcus Count	ND	1.1±0.06a	1.1±0.03a	ND	ND
Yeast Count	2.6±0.01b	2.2±0.03b	2.2±0.07b	2.3±0.02b	3.4±0.07a
Mould Count	1.2±0.01a	2.6±0.21c	3.1±0.05b	2.4±0.02c	3.3±0.02b

Values are means + standard deviations of triplicate determinations. Means with the same superscript with a column are significantly different ($p \le 0.05$)

Table 3: Microorganisms isolated from Pito beverage locally prepared and hawked in five different locations Ogun state southwestern Nigeria.

Samples	Isolates				
Control	Streptococcus sp, Micrococcus sp, Saccharomyce sp, Rhizopus sp, and Mucor sp,				
Abeokuta	Streptococcus sp, Klebsiella sp, Micrococcus sp Staphylococcus sp Salmonella sp. Saccharomyce sp, Aspeergillus sp, Penicillium				
	sp,Sporotrichum sp, Rhizopus sp, and Mucor sp,				
Sango	Streptococcus sp, Klebsiella sp, Micrococcus sp and Salmonella sp. Saccharomyce sp, Aspeergillus sp, Penicillium sp,				
	Sporotrichum sp, Rhizopus sp, Mucor sp, Staphylococcus sp				
IEO	Streptococcus sp, Klebsiella sp, Micrococcus sp and Salmonella sp. Saccharomyce sp, Aspeergillus sp, Penicillium sp,				
IFU	Sporotrichum sp, Rhizopus sp, Mucor sp,				
Agbara	Streptococcus sp, Klebsiella sp, Micrococcus sp and Salmonella sp. Saccharomyce sp, Aspeergillus sp, Penicillium sp,				
Ũ	Sporotrichum sp, Rhizopus sp				

Table 4: Sensory evaluation of Pito beverage locally prepared and hawked in five different locations Ogun state southwestern Nigeria.

Formulas	Parameters					
Samples	Control	Abeokuta	Sango	Ifo	Agbara	
Colour	7.90b	8.90a	8.20a	7.60b	8.20a	
Aroma	7.70b	8.50a	7.50b	6.70c	7.50a	
Taste	8.50a	7.90b	8.20a	7.70b	8.10a	
Overall Acceptablity	8.90a	8.20a	8.70a	7.50b	8.00a	

Means with the same superscript with a column are significantly different ($p \le 0.05$)

Discussion

Proximate Analysis

The result of the Proximate composition of Pito beverage locally prepared and hawked in five different locations in Ogun state Nigeria is shown in Table 1. The moisture, ash, fat, protein and carbohydrate content of the Pito beverage samples are significantly different with values that ranged from 90.12 to 95.42%, 0.74 to 0.93%, 0.16 to 0.32%, 0.41 to 0.58% and 3.21 to 7.58%, respectively. The highest moisture content (95.42%) was recorded in Pito beverage that was freshly prepared while the least (90.12) was recorded in hawked Pito beverage obtained from Ifo, Ogun State. The highest ash content (0.93%) was recorded in Pito beverage that was freshly prepared while the least (0.74%) was recorded in hawked Pito beverage obtained from Agbara, Ogun State. The highest fat content (0.32) was recorded in Pito beverage that was freshly prepared while the least (0.16) was recorded in hawked Pito beverage obtained from Abeokuta, Ogun State. The highest protein content (0.58%) was recorded in Pito beverage that was freshly prepared while the least (0.41%) was recorded in hawked Pito beverage obtained from Agbara, Ogun State. The highest cabohydrate content (8.31%) was recorded in

hawked Pito beverage obtained from Ifo, Ogun State while the least (3.21%) was recorded in Pito beverage that was freshly prepared. The absences of Crude fibre further demonstrated the desirable nutritive quality of the fresh Pito beverage produced. The low protein content of the Pito beverages produced is good for maintenance of cellular organization (Desai and Wagh, 1995)^[7].

Microbiological Analysis

The result of the Microbiological analysis of Pito beverage locally prepared and hawked in five different location in Ogun state Nigeria was shown in Table 2: The Total viable count ranges from 4.1 to 6.3×10^6 cfu/ml, Coliform count ranges from 1.3 to 4.1 x9-

36 10^6 cfu/ml, *Salmonella* Count ranges from 1.3 to 3.2 x 10^6 cfu/ml, *Staphylococcus* Count were not detected in hawked freshly prepared Pito beverages obtained from, Ifo and Agbara in Ogun State. Yeast Count ranges from 2.2 to 3.4 x 106 cfu/ml and Mould Count ranges from 2.4 to 4.2 x 10^6 cfu/ml. Microscopic examination of the sample indicated the presence of microorganisms. The microbes were presented in Table 3, indicated the organisms to be *Streptococcus sp, Staphylococcus* sp, *Klebsiella* sp,

Micrococcus sp and Salmonella sp. And the suspected fungi to be Saccharomyce sp, Aspergillus sp, Penicillium sp, Sporotrichum sp, Rhizopus sp, Mucor sp, The result obtained was in agreement with those reported by Kolawole et al. (2007)^[16]. These micro-organisms isolates are of great concern since most of them are pathogenic to man. The presence of Staphylococcus species in the samples may be attributed poor handling during to production. Staphylococcus species in a normal flora of the body and mucous membrane is linked to aetiological agent of septic arthritis (Ellen and Sydney, 1990)^[9]. The organisms can pass onto the food during harvesting, processing or even storage. The consumer is at risk of acquiring food borne disease.

Staphylococcus species is the major cause of staphylococcal poisoning which is characterized by diarrhea and vomiting (Eze *et al.*, 2008) ^[11]. However, the recommended specifications for beverages served in the Gulf region suggests that the maximum count permitted for total colony count of coliforms, yeast and molds are 1×10^4 and 1.0×10^3 CFU/ml, respectively (Gulf Standards, 2002) ^[13]. This is similar to result and conclusion of Mahale *et al.*, (2008) ^[19] in their study on microbiological analysis of street vended fruit juices from Mumbai city in India.

Sensory Evaluation

The results for the sensory evaluation and overall acceptability of Pito beverage locally prepared and hawked in five different locations in Ogun state Nigeria were Presentedin Table 4. The statistical analysis revealed that there were significant difference (p<0.05) among the fresh and hawked Pito samples in the sensory attributes observed. Hawked pito obtained from Abeokuta, Ogun State had the highest score (8.90), while Hawked Pito obtained from Ifo, Ogun State had the lowest score (7.60) for Colour. Browning in the beverages could have been due to Maillard-type reactions (Potter and Hotchkiss, 2006) ^[21] resulting from the presence of reducing sugars, proteins and amino acids. It may also be due to the effect of severe heating during processing on the quality attributes (Mannay and Shadaksharaswany, 2005) ^[20].

Similar trends were observed for the sensory ratings of Aroma and aroma in the hawked Pito beverages. Acidity contributes to the development of aroma through a proper sugar-acid ratio thereby modifying the sweetness of sugar (Adeola and Aworh, 2010)^[1].

The residual after taste was characterized by perceived bitterness after swallowing the orange juice samples by the panelists. This bitter perception was prominent in hawked Pito obtained from Abeokuta (7.90) and hawked Pito obtained from Ifo (7.70). The panelists affirmed that this was responsible for their low scores on aroma. Mannay and Shadaksharaswany (2005) ^[20]. Reported that the inclusion of additives could impact on the organoleptic qualities of food products. Hawked Pito obtained from Abeokuta, Sango and freshly prepared had the best overall acceptability.

Conclusion

The Hawked Pito beverage investigated in this study had higher microbial load than the specifications set for beverages in some parts of the world. On the basis of the gulf standards, it is clear that the microbial count in the beverages exceeded the standard by considerable margin. These high counts, however, may pose hazard to the health of consumers especially if pathogenic species are present in the beverages to be consumed. Hawked vendors were mostly uninformed of good hygienic practices (GHP) and causes of diarrhea diseases which could increase the risk of street food contamination. They were also unaware of food regulations as well as lacking supportive services such as water supply of good and adequate quality, waste disposal systems which enhance their ability to provide safe food

Recommendation

The government health agencies must adopt measures to educate the vendors on food safety and hygienic practices. Regular monitoring of the quality of beverages for human consumption must also be enforced. There is need to educate the beverage makers and retailers on the hazards associated with the cultivation of nonchalant attitudes to hygienic processing, display and packaging of these beverages. There should, also, be regular training/retraining and health education of handlers in all aspects of food hygiene and safety.

Competing interest

Authors declare no competing exist

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