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MICROBIOLOGICAL QUALITY ASSESSMENT OF COMMERCIALLY PREPARED YOGHURT RETAILED IN SOKOTO STATE, NORTHWESTERN NIGERIA.

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ABSTRACT

Microbiological quality of six brands (18 samples) of yoghurt sampled from retail outlets in Sokoto, North-western Nigeria were evaluated for the level of quality measures used by the producers and their wholesomeness for human consumption.

The mean pH value of the yoghurt samples ranges from 3.69-4.50. The mean total aerobic plate count (TAPC) was 3.37 x 10^4 cfu/ml while the range was 2.66 x 10^4 ± 8.3 x 10^3 to 1.13 x 10^5 ± 1.17 x 10^5cfu/ml of yoghurt. There was no significant difference (P>0.05) in the cfu/ml between the six yoghurt brands analysed.

The aerobic plate mesophilic bacteria isolated were *Lactobacillus* spp and *Streptococcus* spp while E. *coli* an indicator for coliforms was not detected in any of the samples. The findings of this study indicates that the bacterial load of some commercially produced yoghurt sold in Sokoto were within the recommended safe limits of 1.0 – 3.5 x 10^5 cfu/ml for human consumption.

Key words: Aerobic plate count, Coliform, Yoghurt, pH.

INTRODUCTION

Yogurt is one of the most popular fermented dairy products widely consumed all over the world. It is obtained by lactic acid fermentation of milk by the action of a starter culture containing *Streptococcus thermophilus* and *Lactobacillus delbruckii* spp. *bulgaricus*. The role of these two genera in yogurt manufacturing can be summarized as milk acidification and synthesis of aromatic compounds (Serra *et al*., 2009). Yogurt has also been described as a notoriously balanced food containing almost all the nutrients present in milk but in a more assimilable form and they can be produced from skimmed or whole milk and there is a large range of flavours available commercially (Anthar, 1986; Oyeleke *et al*., 2009). People who are moderately lactose intolerant can consume yoghurt without ill-effect because much of the lactose in the milk precursor is converted to lactic acid by the bacterial culture (Alayande, 2008). In Nigeria, it is a popular drink due to its nutritional, probiotic, vulvovaginal and organoleptic characteristics. Fermented milks, like the fresh milk from which they are produced, are liable to microbial contamination and be a risk to human health. Moulds and yeast are the primary contaminants in yogurt produced commercially in Nigeria (Suriyarachchi and Fleet, 1981; Oyeleke *et al*., 2009; De *et al*., 2014).

Though bottled yogurt is a popular drink in Sokoto Metropolis, Sokoto state, Nigeria, limited research has been done to assess the microbiological quality especially in Nigeria.

MATERIALS AND METHODS

Study area

The study was conducted within Sokoto Metropolis, Sokoto State, Nigeria. It is located at the extreme Northwest of Nigeria within the Sudan savannah between Latitude 12^{o}N and $13^{o}58'$N and Longitude $4^{o}8'$E and $6^{o}54'$E.

There were six (6) commercial yoghurt producers known to the investigators within the study area. The study was conducted between August – November, 2011.

Sample collection

Commercially prepared yogurt products packaged in plastic containers were purchased from retailers. A total of eighteen (18) samples were purchased, three samples each from the six (6) available commercial yoghurt producers at a sampling interval of one (1) week each. The information, on their claim forms were recorded e.g., National Agency for Food, Drug, Administration and Control, NAFDAC registration number, manufactured and expiry dates, address of producers and batch number. Samples were immediately transported to the microbiology laboratory of the Faculty of Veterinary Medicine, Usmanu Danfodiyo University, Sokoto, Sokoto State for analysis.

Sample analysis

Data generated was analysed using one way analysis of variance (ANOVA).

Appearance of yoghurt samples

All the samples of yoghurt purchased were homogenous, white in colour, semi solid in nature, taste good, no granules or pebbles, non-ropy, no blood clots and had pH ranging from 3.69 – 4.50.

Isolation, characterization and identification of isolates

Citrate medium, EMB agar, Nutrient agar, MacConkey agar, Urease medium, MR-VP medium and Indole medium were prepared according to the manufacturer's instruction. The samples of the yogurt products were shaken vigorously to suspend the microbial content. These samples were separately inoculated into different culture media, which include MacConkey agar and Nutrient agar.

Serial dilutions up to 10^{-3} were made and 1 ml of each suspension was introduced onto agar medium using streak method. Serial dilution was carried out by pipetting 1ml of the yoghurt sample into a test tube containing 9mls of distilled water to form 10^{1} and allowed to dissolve properly. One ml of the homogenate was aseptically measured into subsequent test tubes containing 9mls of distilled water and dilutions up to 10^{3} were made as described by Cheesbrough, 2002. The plates were incubated aerobically at 37^{0}C for 24 hours. Discrete colonies were isolated and re-inoculated onto appropriate medium in order to obtain pure isolates. These pure isolates were then kept at 4^{0}C for identification purpose (Michaylova, 2007).

Bacterial isolates were characterized based on microscopic appearance, colonial morphology and biochemical tests. The isolates were identified by comparing their characteristics with those of known taxa (Cheesbrough, 2003; Oyeleke and Manga, 2008).

Colonies were counted and plates with less than 10 and greater than 300 colonies were not considered (Cheesbrough, 2003). The colony forming unit (CFU) per ml was obtained by

multiplying the number of colonies on the end point plate by the dilution factor (Sieuwerts *et al*., 2008)

Coliform indicator isolates
E.coli, a coliform indicator was absent in all the samples analysed.

RESULTS

Table 1 showed the results of evaluation of yogurt companies with manufacturing ethics. It showed that all the different brands except sample II had NAFDAC's registration number similarly, they all indicated name and address, volume, manufacturing and expiry dates. Unlike others, sample II did not indicate batch number on the label.

Table 2 showed total aerobic bacterial plate count of all yoghurt samples analysed with a mean aerobic plate count (TAPC) ranging from $2.66 \times 10^4 \pm 8.3 \times 10^3$cfu/ml to $1.13 \times 10^5 \pm 1.17 \times 10^5$cfu/ml. Sample V had the highest bacterial count of 1.13×10^5cfu/ml and sample VI had the lowest bacterial count of 2.66×10^4cfu/ml. There was no significant statistical difference observed in the cfu/ml of the eighteen (18) yoghurt samples analysed P (> 0.05).

Table 3 showed bacterial isolates from yoghurt samples in Sokoto. Aerobic mesophilic bacterial organisms identified were *Lactobacillus* spp. and *Streptococcus* spp. which are also referred to as bacterial cultures used in the fermentation process. *E.coli*, a coliform indicator was not present in all the samples.

Table 1: Physical examination of container for labelling compliance in Sokoto

S/NO	NAME	NAFDAC REG. NO	NAME & ADDRESS	VOLUME	MANUFACTURING DATE	EXPIRY DATE	BATC NO
1	SAMPLE I	+	+	+	+	+	+
2	SAMPLE II	-	+	+	+	+	-
3	SAMPLE III	+	+	+	+	+	+
4	SAMPLE IV	+	+	+	+	+	+
5	SAMPLE V	+	+	+	+	+	+
6	SAMPLE VI	+	+	+	+	+	+

+ = present, **-** = not present, **MAN**. = manufacturing date **EXP**. = expiry date.

Table 2: Total bacterial plate count (TAPC) of yoghurt.

Commercial Producers.

Weekly Samples	I	II	III	IV	V	VI
First week	6.4×10^4	4.8×10^4	9.6×10^4	4.0×10^4	6.0×10^4	2.0×10^4
Second week	4.4×10^4	4.0×10^4	1.2×10^4	3.6×10^4	3.2×10^4	2.4×10^4
Third week	2.4×10^4	8.8×10^4	4.8×10^4	2.24×10^5	2.48×10^5	3.6×10^4
Mean	4.4×10^4	5.86×10^4	5.2×10^4	1.0×10^5	1.13×10^5	2.66×10^4
SD	2.0×10^4	2.5×10^4	4.21×10^4	1.07×10^5	1.17×10^5	8.3×10^3

Table 3: Isolated organisms from yoghurt in Sokoto

Organisms	I	II	III	IV	V	VI
Streptococcus spp	+	+	+	+	+	+
Lactobacillus spp	+	+	+	+	+	+
Escherichia coli	-	-	-	-	-	-

+ = present, - = absent

DISCUSSION

Regulations by NAFDAC, requires that food labelling should be informative and accurate. The minimum labelling requirements for regulated items involve a declaration of the production brand, name, address of the manufacturer, product's batch number, manufacturing and expiry date, volume and most importantly NAFDA registration number. Of the total six (6) brands analysed in this research, five (5) met the requirements of NAFDAC including batch numbers were displayed on the products' container and the remaining one displayed neither NAFDAC registration number nor batch number. Lack of displaying these NAFDAC requirements could be of great public health concern as to the difficulty it will pose in identifying a specific batch found to be harmful should there be an emergency.

The total aerobic plate count often signifies the sanitary condition of food production processes. The result of this study revealed that significant level of sanitary measures are being put in place throughout the process of production by the commercially available yoghurt producers within the study area.

The mean total aerobic plate count detected in this research work ranges from $2.66 \times 10^4 \pm 8.3 \times 10^3$cfu/ml to $1.13 \times 10^5 \pm 1.17 \times 10^5$cfu/ml.

Statistical comparison revealed that the cfu/ml of analysed samples falls within the standard. Thus they were not contaminated and fit for consumption. However, the isolation of Streptococcus spp and lactobacillus spp could be of public health significance.

***Streptococcus* spp**. is a gram-positive, microaerophilic cocci (round) and non-motile organism which occurs in chains or pairs. It is characterized into Groups A, B, C, D, F, and G.

They can cause septic sore throat, diarrhea and other clinical syndromes similar to staphylococcal intoxication and can be transmitted to humans via food e.g. milk etc.

***Lactobacillus* spp.** *is* a rod-shaped, gram-positive, non-spore-forming and non-motile bacteria of the family Lactobacillaceae. Similar to other genera in the family, *Lactobacillus* are characterized by their ability to produce lactic acid as a by-product of glucose metabolism. They

are widely distributed in animal feeds, manure and milk and milk products. Various species of *Lactobacillus* are used commercially during the production of sour milks, cheeses, and yoghurt. They are commensal inhabitants of animal and human gastrointestinal tracts, as well as the human mouth and the vagina and are commercially used as probiotics to restore normal flora after the imbalance created by antibiotic therapy.

Their isolation in the analyzed samples could be a possible threat to humans considering the role they play in causing clinical infections.

Coliforms were not identified in the analysed samples which could be due to the significant level of sanitary measures implored by the various processing procedures (Jervis, 2002). The absence of these organisms in the samples does not mean they will be absent even at the consumer level as there is possibility of contamination along the chain of distribution.

CONCLUSION AND RECOMMENDATION

This study evaluated the amount of sanitary measures employed by commercial yoghurt producers within the study area (Sokoto) by calculating the total aerobic plate count of each sample from the finished products and determining the source of contamination through identification of micro-organisms in the sample(s). This therefore shows that, expected sanitary measures are being put in place because the total aerobic plate count falls within the standard of $1.0 – 3.0 \times 10^5$ cfu/ml (NAFDAC, 2005), thus fit for human consumption.

The samples with the highest and lowest aerobic plate count are 2.66×10^4cfu/ml and 1.13×10^5cfu/ml respectively, and two (2) mesophilic bacterial organisms *Streptococcus* spp. and *Lactobaccilus* spp. were isolated. The result revealed that yoghurt commercially produced in Sokoto is of good quality; in line with Oyeleke (2009) all effort should be made to sustain the standard and existing regulations of NAFDAC in relation to food quality should be strictly adhered to.

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