

## Microbiological and Nutritional Qualities of Dairy Products: Nono and Wara

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**ABSTRACT:** The microbiological and nutritional qualities of two fermented dairy products: nono and wara were investigated. Bacteria and fungi were isolated from both products. The bacteria isolated include *Acinetobacter mallei*, *Alcaligenes faecalis*, *Bacillus cereus*, *Enterobacter aerogenes*, *Enterobacter cloacae*, *Micrococcus spp*, *Serratia spp*, *Flavobacterium spp*, *Staphylococcus aureus* and *Klebsiella licquifasciens*. The fungal isolates were *Aspergillus niger*, *Aspergillus fumigatus*, *Penicillium chrysogenum*, *Rhizopus spp*, *Fusarium moniliforme*, and *Trichoderma reesii*. The mean total plate count of nono was  $3.55 \times 10^8$  cfu/ml, while that of wara was  $4.55 \times 10^8$  cfu/g. The mean coliform count was  $4.25 \times 10^7$  cfu/ml for nono and  $2.40 \times 10^7$  cfu/g for wara. While the mean fungal count was  $12.9 \times 10^6$  cfu/ml for nono and  $1.31 \times 10^7$  cfu/g for wara. The respective moisture content and total titratable acid were higher in nono (86.03%, 1.37%) than in wara (55.68%, 0.48%). Wara was of a higher pH (4.64), than nono (2.87). Wara had higher values respectively for fat, ash, and protein (18.55%, 1.5%, and 23%) than nono (3.68%, 0.97%, and 6.40%), while the carbohydrate level was lower in wara (1%) than in nono (2.9%). Nono and in particular, wara, are of good protein sources. However, the range of microorganisms isolated from both products pose serious threat to food safety, and the need to ensure the microbiological safety of these products can not be over emphasized. [Nature and Science. 2006;4(3):37-40].

**Keywords:** Nono; wara; microbiological and nutritional qualities; food safety

### INTRODUCTION

Nono and wara are local dairy products that are produced and widely consumed in many African countries, including Nigeria. Mainly the Fulanis produce milk locally in Nigeria and the excess milk is processed into these products for preservation (Akinyele *et al.*, 1999). Nono and wara are produced in homes, especially in villages where shelf-life and safety of the products are not considered. They are however sold to both rural and urban people as food. Nono is produced from non-pasteurized cow milk collected in a container called calabash and allowed to ferment naturally for 24 h (Eka and Ohaba, 1977; Olasupo *et al.*, 1996). Wara is a white soft non-ripened cheese made by the addition of a plant extract (*Calotropis procera*) to the non-pasteurized whole milk from cattle (Adeyemi and Umar, 1994).

Poor hygiene, practiced by handlers of these products, may lead to introduction of pathogenic microorganisms into the products and since they do not undergo further processing before consumption, these foods may pose risk to their consumers.

This research is aimed at isolating and identifying microorganisms present in nono and wara in addition to determining the nutritional and some physicochemical

qualities of nono and wara. These will help to ascertain the microbial and product quality of nono and wara.

### MATERIALS AND METHODS

#### Collection of samples

Nono samples were purchased from hawkers at Yaba, Okoko, Idiaraba, Sabo, and Mile 12 markets and the samples were labeled A, B, C, D, and E respectively. While wara samples were purchased from markets in Orile, Oshodi, Mushin, Ketu, and Agege, all in Lagos state, Nigeria, and the samples were labeled F, G, H, I, and J respectively.

#### Determination of the physicochemical parameters of nono and wara

##### Moisture content

A clean platinum dish was dried in an oven and cooled in a desiccator. The cooled dish was weighed. From the nono sample 5 g was weighed and spread on the dish, the dish containing the sample was weighed. It was then transferred into the air oven at  $105^{\circ}\text{C}$  to dry for about 3h. A pair of tongs was used to transfer the dish into a desiccator, allowed to cool and weighed. The dish was returned into the oven for 30 min and again cooled in a desiccator and weighed (AOAC, 1980). The process

was repeated until a constant weight was obtained. This was done for other samples.

### Hydrogen ion concentration (pH)

In order to obtain the pH of the samples, 5 g each was weighed and suspended in 10 ml of distilled water. The pH was determined with a pH meter.

### Nutritional analysis

The carbohydrate was determined according to Anthrone reaction method (Southgate, 1976). Protein was analyzed for by the Microkjedhal estimation of nitrogen, using a conversion factor of %N x 6.25. Determination of the ash content was done according to AOAC (1980) method, while that of fat was done as described by Pearson (1976).

### Determination of crude fiber

The nono and wara samples to be analyzed were defatted using petroleum ether. From the defatted sample 1 g was weighed into 500 ml beaker containing 100 ml of trichloroacetic acid. The content of the beaker was boiled and refluxed for 4 min. It was cooled and filtered with filter paper (Whatman No.1). The residues were washed six times in hot distilled water and once with mentholated spirit. The filter paper together with the sample was transferred into a porcelain crucible and dried in an oven overnight at 100°C, after which it was cooled in a desiccator, weighed, ashed in a muffle furnace at 600°C for 4h and weighed again after cooling. The loss in weight during incineration was equivalent to the amount of crude fiber.

### Microbiological analysis

This was done both quantitatively and qualitatively. Samples were diluted serially using sterile distilled water as diluent. From appropriate dilutions 1ml each was plated in duplicate using the pour plate method.

Media used include nutrient agar, MacConkey agar, and potato dextrose agar. The nutrient and MacConkey agar plates were incubated at 37°C for 24 h, while the potato dextrose agar plates were incubated at 25°C for 48-72 h. After incubation, developed colonies were counted, and representative colonies were sub cultured to obtain pure cultures of isolates.

### Identification of microbial isolates

Identification of the bacterial isolates was based on cultural, morphological, and biochemical characteristics following standard methods (Buchanan and Gibbons, 1974) while that of fungi was also based on cultural and morphological characteristics and standard methods were followed (Talbot, 1971; Bryce, 1992).

### RESULTS

The moisture and total titratable acid were higher in nono than in wara, and the pH of wara was higher than that of nono (Table 1). From the nutritional analysis, wara had higher values for fat, ash and protein than nono. Carbohydrate was however more in nono than in wara. Crude fiber was not detected in both products (Table 2).

The mean total plate count of nono was  $3.55 \times 10^8$  cfu/ml while that of wara was  $4.55 \times 10^8$  cfu/g. The mean coliform count was  $4.25 \times 10^7$  cfu/ml for nono and  $2.40 \times 10^7$  cfu/g for wara. While the mean fungal count was  $12.9 \times 10^6$  cfu/ml for nono and  $1.31 \times 10^7$  cfu/g for wara (Tables 3a and 3b). The bacteria isolated from the products were *Acinetobacter mallei*, *Alcaligenes faecalis*, *Bacillus cereus*, *Enterobacter aerogenes*, *Enterobacter cloacae*, *Micrococcus* spp, *Serratia* spp, *Flavobacterium* spp, *Staphylococcus aureus*, and *Klebsiella licquifasciens* (Table 4). The fungal isolates include *Aspergillus niger*, *Penicillium chrysogenum*, *Rhizopus* spp, *Fusarium moniliforme*, *Trichoderma reesii*, and *Aspergillus fumigatus*.

Table 1. Physicochemical parameters of nono and wara

Parameters	Nono	Wara
Moisture (%)	86.03	55.68
Total titratable acid (%)	1.37	0.48
pH	2.87	4.64

Table 2. Nutritional content of nono and wara

Parameters ( % )	Nono	Wara
Fat	3.68	18.55
Ash	0.97	1.50
Crude fiber	not detected	not detected
Protein	6.40	23.00
Carbohydrate	2.90	1.00

Table 3a. Microbial count of nono

Locations	Total plate count (x 10 <sup>8</sup> cfu/ml)	Coliform count (x 10 <sup>7</sup> cfu/ml)	Fungal count (x 10 <sup>6</sup> cfu/ml)
A	2.65	3.80	9.5
B	1.75	2.10	5.0
C	2.25	5.20	8.5
D	1.80	4.10	18.5
E	9.30	6.00	23.5
Mean	3.55	4.25	12.9

Table 3b. Microbial count of wara

Locations	Total plate count (x 10 <sup>8</sup> cfu/g)	Coliform count (x 10 <sup>7</sup> cfu/g)	Fungal count (x 10 <sup>7</sup> cfu/g)
F	1.50	1.55	1.00
G	10.90	2.65	1.15
H	3.90	2.40	1.90
I	3.50	3.20	1.35
J	3.70	2.20	1.15
Mean	4.55	2.40	1.31

Table 4. Bacteria isolated from nono and wara

Bacterial isolates	Nono	Wara
<i>Acinetobacter mallei</i>	-	+
<i>Alcaligenes faecalis</i>	-	+
<i>Bacillus cereus</i>	+	+
<i>Enterobacter aerogenes</i>	+	-
<i>Enterobacter cloacae</i>	-	+
<i>Micrococcus</i> spp	+	+
<i>Serratia</i> spp	-	+
<i>Flavobacterium</i> spp	+	-
<i>Staphylococcus aureus</i>	+	+
<i>Klebsiella licuifasciens</i>	-	+

Key: + = Present; - = Absent

## DISCUSSION

The nutritional content of nono and wara shows that they are of appreciable nutritional status especially in the protein content. The dairy products particularly wara are good sources of protein. Higher protein content was observed in wara than in nono. This could be due to accumulation of the protein through expulsion of the whey, which does not occur in nono production.

The results obtained from the microbial analysis of nono and wara show that both products were contaminated with microorganisms of public health concern. The high total bacterial and coliform count in both products may be a consequence of the low level of hygiene maintained during the processing and sale of the products. This includes the handlers, quality of water used and the utensils. During the sale of wara, dirty hands and spoons are dipped into the bowl for product selection by both hawkers and consumers. The exposure of both wara and nono while they are displayed for sale in bowls can serve as source of

contamination. The detection of *Enterobacter aerogenes*, *Enterobacter cloacae*, *Klebsiella*, and *Serratia* species in nono and wara as the case may be, indicates possible faecal contamination. Being enteric bacteria, their presence indicates poor hygienic practices among handlers of nono and wara. Due to the significance of the faecal-oral route transmission for many bacterial food-borne diseases, basic hygiene measures assume a decisive importance in food safety management (Untermann, 1998).

Other bacteria isolated include *Acinetobacter*, *Alcaligenes*, *Flavobacterium*, *Micrococcus* species, and *Staphylococcus aureus*. *Alcaligenes* and *Flavobacterium* have been implicated in the spoilage of milk and its products at even refrigerator temperatures. *Acinetobacter* species are known to cause ropiness of milk and secretion of extracellular enzymes both at psychrophilic and mesophilic temperatures (Gilmour and Rowe, 1990).

*Bacillus cereus* which is known to be highly resistant to environmental stress due to its sporing

nature was also isolated, and *B.cereus* is known to be of public health importance since it is pathogenic. The detection of *Staphylococcus aureus* is also of public health importance because of its ability to cause a wide range of infections especially food-borne intoxication. This organism was equally isolated by Olasupo *et al.* (2002) from wara and kunun-zaki, a cereal based, non-alcoholic beverage.

The fungal isolates: *Aspergillus*, *Penicillium*, *Rhizopus*, *Fusarium*, and *Trichoderma* species which were isolated are known spore formers, which therefore means that they can easily contaminate the dairy products which are usually exposed during processing, storage, and hawking. They are major spoilage organisms of carbohydrate foods (Rhodes and Fletcher, 1966). However, their growth can result in the production and accumulation of mycotoxins which are of public health and economic importance.

Microbiological standards have not been put in place for the two locally processed dairy products: nono and wara in Nigeria, and even in most African countries where they are consumed. This is also applicable to many other locally processed foods in Nigeria. The isolation of coliforms and some other food pathogens from these dairy products pose serious threat to food safety, especially locally processed foods in Nigeria. Due to the fact that nono and wara are ready-to-eat foods which are consumed without further processing, great attention should therefore be given to the microbiological safety of these products because their direct consumption may cause health hazard to the consumers.

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