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Bio-Burden and Antibacterial Susceptibility Pattern of Repacked Milk Powder on Sale in Ogun State Markets, South-Western Nigeria.

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Abstract

The bio-burden and antibacterial susceptibility profile of milk powder repacked for sale under market conditions in Ogun State, South-Western Nigeria, was investigated. A total of 12 repacked milk powder samples were procured from 3 main markets in 4 different locations (Abeokuta, Sagamu, Ago-Iwoye and Ijebu-Ode) in Ogun State. Total bacterial, coliform, staphylococcal, *E. coli*, and fungal counts as well as the antibiogram of bacterial isolates were determined using standard microbiological techniques. Several pathogenic bacteria and mycotoxigenic fungi were isolated from the samples. *Staphylococcus aureus* was the most prevalent isolate (50.2%) followed by *E. coli* (21.8%), *Bacillus* spp. (10.0%), *Micrococcus* spp. (6.8%), *Streptococcus* spp. (3.7%), *Pseudomonas* spp. (2.5%), *Salmonella* spp. (2.5%), and *Klebsiella* spp. (2.5%). Among the fungi, *Aspergillus* spp. recorded the highest prevalence (38%), followed by *Mucor* spp. (21%), *Penicillium* spp. (16%), *Fusarium* spp. (15%) and *Rhizopus* spp. (10%). Mean total bacterial counts ranged from $5.4 \pm 0.02 \times 10^5$ CFU/g to $6.3 \pm 0.01 \times 10^5$ CFU/g and mean total fungal counts was in range of $1.6 \pm 0.02 \times 10^2$ to $1.9 \pm 0.02 \times 10^2$. Coliform count ranged from 35 ± 0.23 MPN/g to 92 ± 0.05 MPN/g. Staphylococcal count was in the range of <10 to $3.9 \pm 0.23 \times 10^5$ CFU/g while *E. coli* count ranged from 41 ± 0.21 MPN/g to 07 ± 0.02 MPN/g. Data analyses showed highly significant ($p < 0.05$) difference between mean total viable counts in this study and the specified standards. All the bacterial isolates were susceptible to cefuroxime and ceftriaxone but resistant to cotrimoxazole, gentamycin, nalidixic acid and ampicillin. This practice of repacking powder milk should be discouraged as it predisposes the wholesome

product to contamination with consequent public health risk.

Keywords: Bioburden, repacked milk powder, public health risk, antibiotics, food safety.

Introduction

Milk powders and cream powder are milk products which can be obtained by the partial removal of water from milk or cream. One reason for drying milk is to preserve it. Milk powder has a far longer shelf life than liquid milk and does not need to be refrigerated, due to its low moisture content. Another reason is to reduce its bulk for economy/ease of transportation. Powdered milk and dairy products include such items as dry whole milk, non-fat (skimmed) dry milk, dry butter milk, dry whey products and dry dairy blends (Sabah El Khier and Yaqoub 2009). Milk is a part of daily diet for adults (especially expectant and breast-feeding mothers) and growing children. Milk is highly valued because it provides important source of many of the nutrients required for the proper development and maintenance of the human body (Deeb *et al.*, 2010).

Milk is highly nutritious and serves as an excellent growth medium for a wide range of



microorganisms contaminating it. The microbial quality of milk and other dairy products is influenced by the initial flora of raw milk, the processing conditions, and post-thermal treatment contamination. Microorganisms that can cause spoilage of dairy products include Gram-negative psychrotrophs, coliforms, lactic acid bacteria, yeasts and moulds. Furthermore, various bacteria of public health concern such as *Salmonella* spp., *Listeria monocytogenes*, *Campylobacter jejuni*, *Yersinia enterocolitica*, pathogenic strains of *Escherichia coli* and enterotoxigenic strains of *Staphylococcus aureus* may also be found in milk (Deeb *et al.*, 2010; Rajput *et al.*, 2009). Therefore, the microbiological examination of milk and dairy products cannot be over emphasised. Microbiological quality analyses are crucial for the assessment of quality and safety, conformation with standards and specifications, and regulatory compliance. Pathogenic bacteria of public health significance in milk have been a major concern since the early days of the dairy industry, as many diseases are transmissible via milk products. Traditionally raw or unpasteurized milk has been given more attention as a major vehicle for transmission of pathogens (Deeb *et al.*, 2010; Rajput *et al.*, 2009; Sabah El Khier and Yaqoub 2009).

Milk powders or cream powder is normally packaged aseptically by the manufacturers with minimum or no microbial contamination for public consumption (Sabah El Khier and Yaqoub 2009). However, retailers have now devised means of repackaging them in different materials for sale in open markets, perhaps due to the economic situations in many developing countries and especially in Nigeria. This practice of repacking powdered milk presents a potential for contamination of the product at market places. These unhygienic practices may lead to microbial contamination due to deposition of bio-aerosols on exposed products and transfer of microbes from dirty hands (Kawo and Abdulmumin, 2009; Adesiyun, 1984). Simple packaging or repackaging operations can bring about favourable condition(s) for the contamination or re-

contamination of the product(s) with pathogens if strict aseptic conditions are not adhered to (Kawo and Abdulmumin, 2009). Produce receive most of their contamination from their ingredients, although some contaminants may be added by unwrapped/unpackaged pieces by air, dust and handling. Further contamination may ensue from contaminated equipment when they come in contact with food during packaging/repackaging and from individual personnel (Kawo and Abdulmumin, 2009; Shamsuddeen *et al.*, 2008; Shamsuddeen and Ameh, 2008). Poor sanitary conditions and environments that are highly impacted with spoilage and pathogenic microbes could generally be the source of contamination of food products exposed to it. Thus, retailers of food products have been implicated in the spread of food-borne diseases (Oyeyi and Lum-nwi, 2008; Shamsuddeen *et al.*, 2008).

Little or no attention has been given to the microbial burden (bacterial and fungal) and public health risks associated with repacked milk powder under market conditions in Ogun State, and Nigeria at large. Therefore, the objectives of the present investigation were to evaluate the microbial burden and safety risks associated with repacked milk powder in Ogun State and assess the antibiotic susceptibility pattern of bacterial isolates.

Materials and Methods

Sample collection

A total of twelve (12) samples, three samples each of repacked milk powder displayed in the open for sale in two different packaging materials (transparent polyethylene bag and plastic container) were purchased from main markets in four (4) different locations (Abeokuta, Sagamu, Ago-Iwoye and Ijebu-Ode) in Ogun State. Samples were randomly and aseptically collected into a 150ml-capacity, wide-mouthed glass containers with air-tight fitting caps, which were previously disinfected with ethyl alcohol and autoclaved at 121°C for 15 minutes. The samples were immediately transported to the laboratory for analyses.

Microbiological analyses

Preparation of samples and inoculation of media

The protocol of Houghty *et al.* (1992) and Christen *et al.* (1992) were adopted. Twenty-five grams (25g) of the repacked milk powder sample was aseptically weighed and introduced into 225 ml of previously-sterilized buffered peptone water (LAB M). This was achieved by aseptically opening of the repackaging materials (by wiping the tied mouth/cover of the polyethylene bags or plastic containers, respectively with ethyl alcohol). Control sample (milk powder in its manufacturer's original packaging material) was also subjected to similar treatment. The mixture (solution) was homogenized by shaking vigorously to produce the homogenate. Subsequently, serial 10-fold dilutions of samples were prepared and 0.1ml aliquots were spread plated on appropriate media. Aerobic plate counts were evaluated on plate count agar (PCA, Oxoid CM 325, Oxoid, UK) using the pour plate method. The most probable number or multiple tube technique was used to detect the presence of both total and faecal coliform. Enterobacteriaceae were counted on violet red bile glucose agar (VRBG, Oxoid CM 485, Oxoid, UK) incubated at 37°C for 24 hours. *E. coli* was enumerated on eosin methylene blue (EMB, Oxoid) agar incubated at 37°C for 24 hours. Staphylococci population was enumerated on mannitol salt agar (MSA, Oxoid) incubated at 37°C for 24 hours. Fungal count was determined using acidified (pH 3.5) potato dextrose agar (PDA, Unipath, Basinstoke Ltd, UK) for pour plating and incubated at 27°C for 3-5 days. All plates were in duplicate.

Characterization and identification of isolates

Characterization and identification of the isolated bacteria were carried out based on their colonial, morphological and biochemical characteristics with reference to the Bergey's Manual of Systematic Bacteriology (Holt *et al.*, 1994). The fungal isolates were identified according to the protocol of Samson and van Reenen-Hoekstra (1988) and which was based on microscopic examination of their conidial heads, phialids, conidiophores and presence or

absence of rhizoids.

Antibiotic susceptibility testing

Antibiotic susceptibilities of bacterial isolates were determined according to the method recommended by the Clinical and Laboratory Standards Institute (2007). Briefly, inocula were prepared for each bacterial isolate by adjusting the turbidity to 0.5 McFarland standard and spread on Muller-Hinton agar plates. Antibiotic discs (Himedia, Mumbai, India) were placed on the agar plates and incubated overnight at 37°C for 24 hours. The zones of inhibition were measured and the isolates were classified as sensitive, moderately sensitive, intermediate, and resistant according to CLSI tables and guidelines. The following antibiotics with the disc strength in parentheses were used: Ciprofloxacin (5µg), Gentamycin (10µg), Nalidixic acid (30µg), Cefotaxime (30µg), Cefuroxime (30µg), Azithromycin (15 µg), Imipenem (10µg), Cephalexin (30µg), Ceftriaxone (30µg), Cotrimoxazole (25µg), Ampicillin (10µg), Erythromycin (15µg). A control strain of *E. coli* ATCC 25922 was included in each plate.

Statistical Analysis

The statistical package used should be included: Data obtained were subjected to statistical analysis using SPSS version 16.0 software to determine means and standard deviations of means. Significant differences between means of experiments were determined by analysis of variance (ANOVA). A significance level was set as $p \leq 0.05$.

Results

Total and mean total aerobic bacterial and fungal counts

The mean and total aerobic bacterial and fungal counts of repacked milk powder samples are indicated in Table 1. It shows that the mean total aerobic bacterial count of the samples ranged from $5.4 \pm 0.02 \times 10^5$ CFU/g to $6.3 \pm 0.01 \times 10^5$ CFU/g while the mean total aerobic fungal counts ranged from $1.6 \pm 0.02 \times 10^2$ to $1.9 \pm 0.02 \times 10^2$.



Table 1: Total and mean bacterial and fungal counts (CFU/g) of repacked milk powder samples from different locations in Ogun State

Location	Bacterial				Fungal			
	S1	S2	S3	Mean T	S1	S2	S3	Mean T
Abk	6.9±0.03x10 ⁵	6.3±0.1x10 ⁵	5.7±0.01x10 ⁵	6.3±0.01x10 ⁵	1.5±0.01 x 10 ²	1.8±0.02 x 10 ²	2.5±0.02 x10 ²	1.9±0.02x10 ²
Sag	5.8±0.01x10 ⁵	5.4±0.1 x10 ³	5.9±0.02x10 ⁵	5.7±0.02x10 ⁵	1.6±0.01 x 10 ³	1.9±0.03 x 10 ³	1.5±0.01 x10 ²	1.6±0.02x10 ²
AgI	5.2± 0.03x10 ⁵	6.1±0.03x10 ⁵	4.9±0.03x10 ⁵	5.4±0.02x10 ⁵	2.0±0.02 x 10 ²	2.1±0.02 x 10 ²	1.7±0.02 x10 ²	1.9±0.02x10 ²
IjO	6.1±0.01x10 ⁵	5.3±0.21x10 ⁵	5.0±0.02x10 ⁵	5.5±0.02x10 ⁵	1.8±0.01 x 10 ²	2.2±0.02 x 10 ²	1.3±0.01 x10 ²	1.7±0.02x10 ²
Control	-	-	-	-	-	-	-	-

Keys: Abk= Abeokuta; Sag= Sagamu; AgI= Ago-Iwoye; IjO= Ijebu-Ode; S1-S3= Samples 1, 2, 3; CFU/g= colony forming unit per gram; values are mean ± standard deviation of two (2) replicates; mean T= Mean of total aerobic counts (S1+S2+S3); - = No growth.

Total coliform, staphylococcal and *E. coli* counts

The highest coliform bacterial count was found in sample 1 from Abeokuta (92±0.05 MPN/g) whereas; sample 1 from Ago-Iwoye market recorded the lowest total coliform count of 35±0.23 MPN/g (Table 2). Staphylococci were found in all the 12 tested samples. The highest staphylococcal count was found in sample 1 from Abeokuta market (3.9±0.23x10⁵ CFU/g)

while samples 2 and 3 from Sagamu and Ijebu-Ode markets both recorded insignificant staphylococcal counts (<10). *E. coli* was isolated from 11 repacked powder milk samples out of 12 samples tested. The highest *E. coli* count was found in sample 2 from Sagamu (41±0.21 MPN/g) while the lowest *E. coli* count (07±0.02 MPN/g) occurred in sample 3 from Abeokuta. *E. coli* was not detected in S2 from Abeokuta (Table 2).

Table 2: Total Coliform, Staphylococcal and *E. coli* counts of repacked milk powder from different locations in Ogun State.

Location	Total Coliform Count (MPN/g)	Total <i>Staphylococcal</i> Count (cfu/g)	<i>E. coli</i> count (MPN/g)
Abe			
S1	92±0.05	3.9±0.23x10 ⁵	21±0.03
S2	70±0.03	3.6±0.12x10 ⁴	-
S3	61±0.04	<30	07±0.02
Sag			
S1	84±0.01	1.50.11±x10 ⁵	28±0.33
S2	64±0.02	<10	41±0.21
S3	45±0.04	4.3±0.02x10 ⁴	23±0.02
AgI			
S1	35±0.23	<30	9.9±0.06
S2	75±0.04	3.2±0.33x 10 ²	6.7±0.31
S3	60±0.33	4.8±0.05x10 ³	15±0.03
IjO			
S1	64±0.01	2.4±0.06x10 ⁴	33±0.22
S2	110±0.03	1.8±0.38x10 ²	38±0.05
S3	40±0.03	<10	7.3±0.23

Keys: Abk= Abeokuta; Sag= Sagamu; AgI= Ago-Iwoye; IjO= Ijebu-Ode; S1-S3= Samples 1, 2, 3; MPN/g= Most probable number per gram; CFU/g= colony forming unit per gram; values are mean ± standard deviation of two (2) replicates; - = not detected.

Percentage prevalence of isolates

The percentage prevalence of bacterial (Figure 1) and fungal (table 3) isolates from the samples revealed that *Staphylococcus aureus* was the most prevalent pathogen (50.2%) followed by *E. coli* (21.8%), *Bacillus* spp. (10.0%), *Micrococcus* spp. (6.8%), *Streptococcus* spp.

(3.7%), *Pseudomonas* spp. (2.5%), *Salmonella* spp. (2.5%), and *Klebsiella* spp. (2.5%). Also, *Aspergillus* spp. recorded highest prevalence (38%), followed by *Mucor* spp. (21%), *Penicillium* (16%), *Fusarium* spp. (15%) and *Rhizopus* spp. (10%) (Table 3).

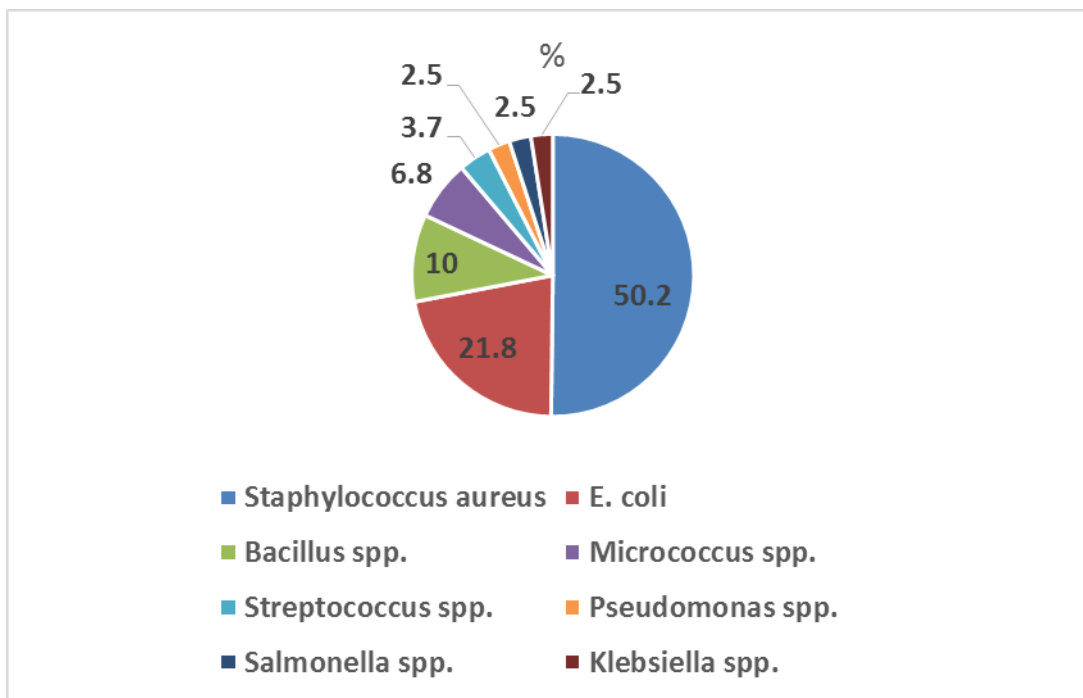


Figure 1: Percentage prevalence of bacterial isolates from repacked milk powder in Ogun State, Nigeria

Table 3: Percentage prevalence of fungal isolates from repacked milk powder in Ogun State, Nigeria

Isolates	Percentage (%)
<i>Penicillium</i> spp.	16.0
<i>Aspergillus</i> spp.	38.0
<i>Mucor</i> spp.	21.0
<i>Rhizopus</i> spp.	10.0
<i>Fusarium</i> spp.	15.0

Antibiotic susceptibility test

All the bacterial isolates were susceptible to cefuroxime and ceftriaxone but resistant to cotrimoxazole, gentamycin, nalidixic acid and

ampicillin. Various results of resistance, intermediate, susceptible and moderate susceptibilities were observed with the other test antibiotics (Table 4).

Table 4: Antibiotic susceptibilities of bacterial isolates from repacked milk powder in Ogun State, Nigeria

Antibiotics content	Disk	Bacterial isolates							
		<i>E. coli</i>	<i>Pseudomonas</i> spp.	<i>Klebsiella</i> spp.	<i>Salmonella</i> spp.	<i>Bacillus</i> spp. Zone diameter (mm)	<i>Micrococcus</i> spp.	<i>Staphylococcus Aureus</i>	<i>Streptococcus</i> spp.
Cotrimoxazole	25 µg	7 (R)	8(R)	5(R)	0	0	12(R)	11(R)	0
Gentamycin	10 µg	13(R)	0	0	0	0	1R(S)	13(R)	15(I)
Nalidixic acid	30 µg	10(R)	0	0	0	0	0	0	0
Ampicillin	10 µg	0	0	0	0	0	17(R)	11(R)	0
Erythromycin	15 µg	15(I)	12(R)	17(S)	0	0	12(R)	13(R)	14(I)
Azithromycin	15 µg	21(S)	22(S)	26(S)	5(R)	12(R)	22(S)	24(S)	0
Imipenem	10 µg	25(S)	20(S)	18(S)	0	10(R)	25(S)	28(S)	21(S)
Ceftriaxone	30 µg	28(S)	23(S)	23(S)	28(S)	23(S)	32(S)	39(S)	19(S)
Cefotaxime	30 µg	21(S)	0	20(S)	18(MS)	0	21(S)	25(S)	17(MS)
Cefuroxime	30 µg	23(S)	22(S)	27(S)	24(S)	25(S)	29(S)	38(S)	24(S)
Cephalexin	30 µg	17(I)	14(R)	21(S)	0	0	20(S)	21(S)	10(R)
Ciprofloxacin	5 µg	24(S)	19(S)	30(S)	23(S)	13(R)	27(S)	26(S)	10(R)

Keys: µg= Microgram; mm= Millimetre; R= Resistant; I= Intermediate; S= Susceptible; MS= Moderately Susceptible; 0= No zone of inhibition

Discussion

Total viable count is an important criterion for assessing the microbiological quality and the degree of freshness of various foods. In this study, mean total bacterial count of repacked milk powder sample ranged from $5.4 \pm 0.02 \times 10^5$ CFU/g to $6.3 \pm 0.01 \times 10^5$ CFU/g. Mean total fungal counts ranged from $1.6 \pm 0.02 \times 10^2$ to $1.9 \pm 0.02 \times 10^2$. Coliform count ranged from 35 ± 0.23 MPN/g to 92 ± 0.05 MPN/g. Staphylococcal count was in the range of <10 to $3.9 \pm 0.23 \times 10^5$ CFU/g while *E. coli* count ranged from 41 ± 0.21 MPN/g to 07 ± 0.02 MPN/g. Coliform count was highest in sample 1 from Abeokuta (92 ± 0.05 MPN/g) with sample 1 from Ago-Iwoye market recording the lowest (35 ± 0.23 MPN/g). Sample 1 from Abeokuta market also recorded highest staphylococcal count ($3.9 \pm 0.23 \times 10^5$ CFU/g) whereas, samples 2 and 3 from Sagamu and Ijebu-Ode, respectively, had insignificant staphylococcal counts (<10). The highest *E. coli* count occurred in sample 2 from Sagamu (41 ± 0.21 MPN/g) while the lowest *E. coli* count (07 ± 0.02 MPN/g) occurred

in sample 3 from Abeokuta. The variations in microbial composition of the samples in this study with regards to location may be due to the level of unhygienic environmental conditions and handling of the repacked milk powder and, their multiplication would however, depend on favourable environmental conditions like time, temperature, relative humidity and duration of storage as well as food parameters such as potential water activity (aW), moisture content, nutrients present (Oyeyipo *et al.*, 2012; Sabah El Khier and Yaqoub 2009; Ahmed and Anwar 2006). The microbial counts in this work showed high deviations from the standard limits specified by the Codex Alimentarius Commission (CAC) (2×10^5 CFU/g) for total viable bacterial count in milk or cream powder. Yeast and mould count should be less than Log_{10} 1.0 CFU/g, and the milk powder must be free from coliform bacteria (SDS, 2008; EAS, 2006; SVGNS, 2004; CAC, 1999). This high microbial load is an indication of contamination/recontamination, since milk powder normally complies with universally acceptable microbiological standards (Deeb *et*



al., 2010; Sabah El Khier and Yaqoub 2009). Analysis of data revealed highly significant ($p < 0.05$) difference between mean total bacterial counts in this study and the specified standards. Similarly, high total viable bacterial and coliform counts have been reported in milk powder under market conditions in other countries (Deeb *et al.* 2010; Rajput *et al.*, 2009; Sabah El Khier and Yaqoub 2009; Ahmed and Anwar 2006).

Eight bacteria genera (*Staphylococcus aureus*, *E. coli*, *Bacillus* spp., *Micrococcus* spp., *Streptococcus* spp., *Pseudomonas* spp., *Salmonella* spp. and *Klebsiella* spp.) were isolated from the repacked milk powder samples while the fungal isolates were: *Aspergillus* spp., *Mucor* spp., *Penicillium* spp., *Fusarium* spp. and *Rhizopus* spp. Majority of these microbes are potent food poisoning and intoxication agents (Adams and Moss, 1995). Many researchers (Kawo and Abdulmumin, 2009; Aboloma, 2008; Oyeyi and Lum-nwi, 2008; Shamsuddeen *et al.*, 2008; Shamsuddeen and Ameh, 2008) have reported that the presence of *Staphylococcus* and *Streptococcus* spp. were possible contaminants from handlers. *Streptococcus* spp. has been implicated in human infections like pharyngitis, scarlet fever and pneumonia. *Staphylococcus aureus*, a mesophile have been implicated in food poisoning outbreak of some food material. Of special interest is the ability of *S. aureus* to elaborate enterotoxins in foods, which is dangerous to human and other animal health (Wieneke *et al.*, 1993). The isolation of *Bacillus* spp. may be attributed to the presence of their spores in the environment. *Bacillus* species are spore formers whose spores could survive high temperatures of processing. They are able to inhabit soils and vegetation and have been repeatedly isolated in several countries from wide variety of foods (Oyeyi and Lum-nwi, 2008; Shamsuddeen *et al.*, 2008; Amusa *et al.*, 2005; Osuntogun and Aboaba, 2004). Their occurrence in the repacked milk powder samples in this study is therefore not surprising because of the manner in which the milk powder is handled in the open market in a dusty and muddy environment. *Bacillus* spp. causes some toxin mediated diseases rather than infections (Adebayo-Tayo *et al.*, 2009) such as diarrhoea

and emetic illness characterized by nausea and vomiting. The infectious dose has been estimated to be $10^5/g$ (Falegan and Oluwaniyi, 2015; Adeleke and Odelola, 1997). As at the time of this study, there was no specification set by the CAC or the ICMSF for *Bacillus* spp. in milk powder, however, the presence of high number of *Bacillus* spp. in repacked powder milk in this study indicates pre/post repackaging (unhygienic handling) contamination as their presence is often used as a post-processing hygienic indicator by most manufacturers. The distribution of enteric bacteria (*E. coli* (21.8%), *Salmonella* spp. (2.5%) and *Klebsiella* spp. (2.5%) in this study agrees with the findings of Falegan and Oluwaniyi (2015) and, may suggest that the repacked milk powder samples have been contaminated with various intestinal bacteria as a result of unhygienic handling on the part of retailers and the repackaging materials. The presence of highly pathogenic bacteria isolates such as *Salmonella* spp., *E. coli*, *Klebsiella* spp. *Pseudomonas* spp., *Bacillus* spp. and *S. aureus* are of concern because of the public risk associated with the consumption of contaminated repacked milk powder. The presence of species of *Aspergillus*, *Penicillium*, *Mucor*, *Rhizopus*, *Fusarium*, could be attributed to the prevalence of their spores in the atmosphere. Fungal spores are found in the air, the spores must have contaminated the milk powder during repacking. Several researchers have reported that liberated spore can easily settle on food and ceilings of room and then germinate (Oyeyipo *et al.*, 2012; Adebayo-Tayo *et al.*, 2009). Most fungi that contaminate food often produce secondary metabolite known as mycotoxin in them which are known to be hazardous to humans. Mycotoxins are naturally occurring toxic chemical compounds which are capable of inducing mycotoxicoses (toxic syndromes especially cancer) in humans following ingestion or inhalation. However, they differ their degree and manner of toxicity (Oyeyipo *et al.*, 2012; Adebayo-Tayo *et al.*, 2009; Ahmed and Anwar 2006). Some of these metabolites include: vomitoxin by *Fusarium* species, ochratoxin A, produced by *Penicillium* species and aflatoxin produced by *Aspergillus* species. These fungi have been detected in food

under market conditions (Oyeyipo *et al.*, 2012; Sabah El Khier and Yaqoub 2009; Ahmed and Anwar 2006). All the organisms isolated in this study have health implication for man except *Micrococcus* spp., which have not been associated with human infections (Falegan and Oluwaniyi, 2015; Oyeyipo *et al.*, 2012; Adebayo-Tayo *et al.*, 2009).

All the bacterial isolates were susceptible to cefuroxime and ceftriaxone but resistant to cotrimoxazole, gentamycin, nalidixic acid and ampicillin. Varied results of resistance, intermediate, susceptible and moderate susceptibilities were observed with the other test antibiotics (table 4). Multiple antibiotics resistance (MAR) has been widely reported in bacteria (Klein *et al.*, 2007; Okuma *et al.*, 2002; Scott, 2002; Allen and Cowan, 1997; Foster, 1996). Multiple antibiotics resistance index is a tool that reveals the spread of resistance in a given microbial population; it implies that the strains of such bacteria originate from an environment where several antibiotics are used. Resistance to antimicrobial agents is a public health threat and increasingly becoming a global problem. It limits therapeutic options and leads to increased mortality and morbidity. Infections with antibiotic resistant bacteria make the therapeutic treatment against infection extremely difficult or virtually impossible in some instances (Falegan and Oluwaniyi, 2015).

Conclusions and Recommendations

This research revealed the presence of pathogenic/toxigenic bacteria and fungi in repacked milk powder on sale in Ogun State markets, South-Western, Nigeria. Microbial counts were generally above the allowable limit in the repacked milk powder to be marketed for consumption. These high counts, in addition to the development of multiple antibiotic resistance are major public health threat to consumers, especially children and the elderly. When antimicrobial drugs are administered on them during illness, infections may be prolonged and may even lead to increase in mortality. Therefore, this practice should be completely discouraged so that only brands of powdered milk that are aseptically processed by

manufacturers, in original packaging materials (tins or bags) and approved by regulatory bodies in Nigeria are sold to consumers.

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