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BACTERIOLOGICAL EVALUATION OF NIGERIAN CURRENCY NOTES FROM SELECTED HANDLERS IN ILESHA METROPOLIS OF OSUN STATE, NIGERIA

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ABSTRACT

Background: Peoples from various background and from different works of life with different hygienic status always engaged in physical transactions with a legal tender of varied denominations for exchange of goods and services, and one of the legal tender officially recognized in Nigeria is called naira. This study evaluated bacterial contaminants onnaira notes in circulations from selected handlers with specific profession and determined the resistance of the isolates to conventional antibiotic in use.

Methods: A total of 160 samples of currency notes 20 each of 8 existing denominations in Nigeria, collected from selected participants of various professions soaked in ringer's solution were serially diluted, subculture to various bacteriological media, Gram stained and biochemically characterized. Determination by antibiogram study was carried out,with a view to identifying resistance risk factors that could be associated with these contaminatedcurrency notes.

Results: The microbial load was found to be higher in lower denominations irrespective of their polymer status. The total bacterial count per milliliter varied between 2.28×10^4 and 4.20×10^7 CFU, while the percentage distributions of isolates; *Staphylococcus aureus* (36.8%), *Escherichia coli* (31.5%), *Bacillus spp* (3.7%) and *Pseudomonas aeruginosa* (27.5%) and varied resistance to antibiotics used were recorded.

Conclusion: Bacterial antibiotic resistance has been associated with treatment failure, high health cost burden and loss of manpower hours due to over hospitalization. The microbial contaminant loads capable of causing opportunistic infection were found to be present in currency notes examined. The alarming resistance of bacteria to selected conventional antibiotics used in this study, serves an indication of potential threat of contaminated currency notes to public health.

1. Introduction

The orientation of people of various occupational background towards unhygienic and rough handling of currency notes in Nigeria can easily promote a safe haven for the growth of health threatening microbial contaminant. Money, a legal tender that is used in exchange for goods and services is handled daily by individuals of different occupational background and socio-economic strata for transaction, notwithstanding the cashless policy that encourage the use of credit card for making payment and the use of automated teller machines for withdrawal and transfer of large volume of cash with convenience[1].

It is rife in our nation to find old dirty, worn-out, and mutilated naira notes around. Aside from the fact that the currency notes have their limited life span and are supposed to be withdrawn at regular interval, poor handling habit of currencies is mainly responsible for this observation and such includes squeezing the currency notes; keeping money in the brassier, dirty apron pockets, handkerchiefs, capping and head-tying the currency on heads, counting currency notes with saliva, tucking the currency notes in dirty socks, keeping currency under the rugs or carpets, simultaneously handlings of bloody meat, palm oil and watery materials during trading. During, partying currency notes are sprayed on people as a token of pride or appreciation, and some of the notes lying on the floor are trampled[2].

Studies have established the occurrence of bacteria, fungi and eggs of parasite on paper currency in many developing nations. In the United State of America, swabs of currencies collected at random from medical personnel and workers in a hospital, when cultured revealed they were contaminated with pathogenic bacteria[3].

Microbes are ubiquitous and are found on the surface of animate and inanimate objects, including money. Research carried out on microbial contamination in Bangladesh showed coliforms and other isolates of enteric origin in 80% of the thirty old two taka notes and from 94% of one dollar bills,*Escherichia coli, Staphylococcus aureus, Pseudomonas species* and *Bacillus species* were isolated[4]

Microbial contaminants could emerge from various sources such as the enteric, skin flora and wounds, aerosol produced when people sneezed, droplets from coughing, animal fur or droppings or wastes. Others could come from the bank's money counting machine. Between the late 1800s and early 1900s, scientists postulated the association of handling money with disease transmission. Subsequently, by modern scientific techniques, these postulations confirmed that pathogenic organisms can be isolated from currency/money surfaces. For example, *Citrobacter* spp., *Escherichia coli, Mycobacterium* spp., *Pseudomonas aeroginosa, Salmonella* spp., and *Staphylococcus aureus*, are among the examples of foodborne pathogenic microorganisms reported on currency notes [5].

Clinical isolates obtained from currency notes are capable of interfering with immune status of the handlers and cause infection of epidemiological magnitudes that could be contagious and hence, economic loss. Variations in species of organisms encountered in previous studies seem to have been distributed based on the materials, country, climate and age[6]. In the context of possibility of contamination of currency notes via the chains of moving from hand to hand of peoples of different occupational background for specific purpose of transaction, this study therefore aimed at assessing the degree of bacterial contamination of Nigerian currency notes obtained from different professionals in Ilesha metropolis, Osun state, Nigeria and determining the antibiogram in relation to resistance obtained.

2. Materials and Methods

2.1 Materials

Paper based naira notes of varied denominations (N5, N10, N20, N50, N100, N200, N500 and N1000), 20% ringer's solution (Sigma Aldrich, Germany), laboratory resins (Pyrex), nutrient agar (Oxoid, UK) , mannitol salt agar(Oxoid, UK) and cetrimide agar(Oxoid.UK)

2.2 Collection of samples, size, location and study units

This cross-sectional study was carried out in Ilesha, a conglomeration of many towns located within the Latitude 7^0 37'40.40N and Longitude 4^0.44'29'.80E coordinates, southwest Nigeria.

A total of one hundred and sixty samples of 8 Nigerian currency notes denominations (N5, N10, N20, N50, N100, N200, N500 and N1000) were randomly collected from mechanics, butchers, beggars, tricyclist, traders and fuel station attendants in Ilesha, Osun state.

A total of 120 peoples comprises 78 males and 42 females of different genders and occupations; mechanics, butchers, beggars, tricyclist, herbs sellers and fuel station attendants with carefree orientation in handling currency were randomly sampled designated as representative units - a microcosm of a macrocosm.

2.3 Bacteriological isolationand identification from the currency notes

Each naira notes with unpleasant smelling, visibly dirty and

overused was soaked in 100mL of sterile Ringer's solution for about 45 minutes, thereafter, incubated at 25±3^{0}C with gentle agitation to detach the microbial loads to the aliquot of the solution. Ten-fold serial dilutions of each aliquot were prepared. A volume of 1mL aliquot of the 10^{-3} dilution was then inoculated by surface spreading method separately onto sterile plates containing nutrient agar, cetrimide nutrient agar, mannitol salt agar and left to dry. The agar plates were inverted and incubated at 37°C for 24 hours.

Based on colonial and cultural characteristics, distinct colonies from the plates were taken, Gram-stained and subjected to biochemical tests including catalase test, oxidase test, coagulase test and indole test. Identified isolates were transferred onto agar slants and kept in the refrigerator at 4°C.

2.4 Determination of total aerobic plate count: TPC

A volume of 10 mL of four- fold serially diluted peptone broth solutionwas added to 90 mL of normal saline, from which 1 mL of resulting solution was diluted to 10^4 and 0.1 mL was inoculated by surface spreading methods separately on to sterile standard plate counts agar media in duplicate set. The plates were incubated at 37^{0}C for 24hrs, and thereafter counted using Quebec's colony counter (Equation 1).

$$\text{Colony forming units} = \frac{\text{number of colonies} \times \text{dilution factor}}{\text{Volume of the culture plated.}}$$

…… Equation 1

2.5 Antibiotic susceptibility testing

Antimicrobial susceptibility profiles of the bacterial isolates were determined using the agar diffusion method of Kirby Bauer[7]. Three to five colonies of the overnight culture of the strains were inoculated into a tube containing tryptone soy broth and were incubated for 24hours at 37^{0}C. The inoculums were standardized by adjusting the broth cultures until the turbidity matched the 0.5 McFarland standards. A sterile cotton swab was dipped into the standardized suspension, drained, and used for inoculating 20 mL of Mueller Hinton agar (Oxoid, UK) on a 100-mm disposable plate. The inoculated plates were aseptically air-dried for 30 minutes, and antibiotic discs were impregnated on the agar plates using flamed forceps. The discs were gently pressed on the agar medium to ensure maximum contact. Discs containing the following antibiotics were used: ceftazidime (30µg), cefuroxime (30µg), gentamicin (10µg), cefixime (5µg), ofloxacin (5µg), Augmentin® (30µg), nitrofurantoin (300µg) and ciprofloxacin (5µg). The plates were incubated aerobically at 37^{0}C for 24 hours before measuring diameters of the zones of inhibition. Sensitive, intermediate, and resistant strains were marked S, I and R respectively as standardized by CLSI[7].

2.9 Statistical analysis

Microsoft Excel® was used to collate the data while descriptive statistics were used to analyze the data.

1. Results

A total of 120 participants of varied age, gender and occupations that were known for frequent hand to hand transaction with currency notes were sampled, the gender distribution varied in relation to profession as shown in Table 1.

Table 1. Demographic identities of the target population

Target population	Gender		Age Range
	Male	Female	
Mechanics	20	0	30-50
Butchers	20	0	30-45
Beggars	8	12	30-60
Motor cyclist	18	2	20-35
Traders	4	16	30-50
Fuel station attendants	8	12	25-35
Total Gender	78	42	

A total denomination of 8 existing currencies were sampled, The average bacterial counts slightly varied from one currency to another as shows in Table 2.

Table 2. Average bacterial total plate counts from different denominations.

Denominations ₦(Naira)	Bacterial count (CFU/mL)
5	$4.20x10^7$
10	$4.35x10^5$
20	$3.45x10^4$
50	$4.20x10^3$
100	$6.22x10^5$
200	$5.10x10^4$
500	$3.25x10^3$
1000	$2.28x10^4$

Four different isolates of bacteria species were obtained from the samples after presumptive biochemical confirmation, which include; *Escherichia coli, Pseudomonas aeruginosa. Staphylococcus aureus* and *Bacillus spp. Bacillus spp* was the least encountered (3.7%), while *Staphylococcus aureus* had the highest prevalence (36.8%), *Escherichia coli* had a prevalence of 31.5% while *Pseudomonas aeruginosa* (27.5%) of the 160 currency notes sampled as shown in Table 3

Table 3. Percentage frequency of the isolates from the currency samples.

Denomination	Number of samples	*Escherichia coli*	*Staphylococcus aureus*	*Pseudomonas aeruginosa*	*Bacillus spp*	Total
₦5	20	14	13	*13*	0	40
₦10	20	16	11	*12*	0	39
₦20	20	13	13	12	4	42
₦50	20	11	16	13	2	43
₦100	20	8	18	9	0	35
₦200	20	10	12	*5*	2	29
₦500	20	8	9	*6*	2	25
₦1000	20	5	7	4	0	16
Total %	160	85 (31.5%)	99 (36.8)	74 (27.5)	10 (3.7)	269

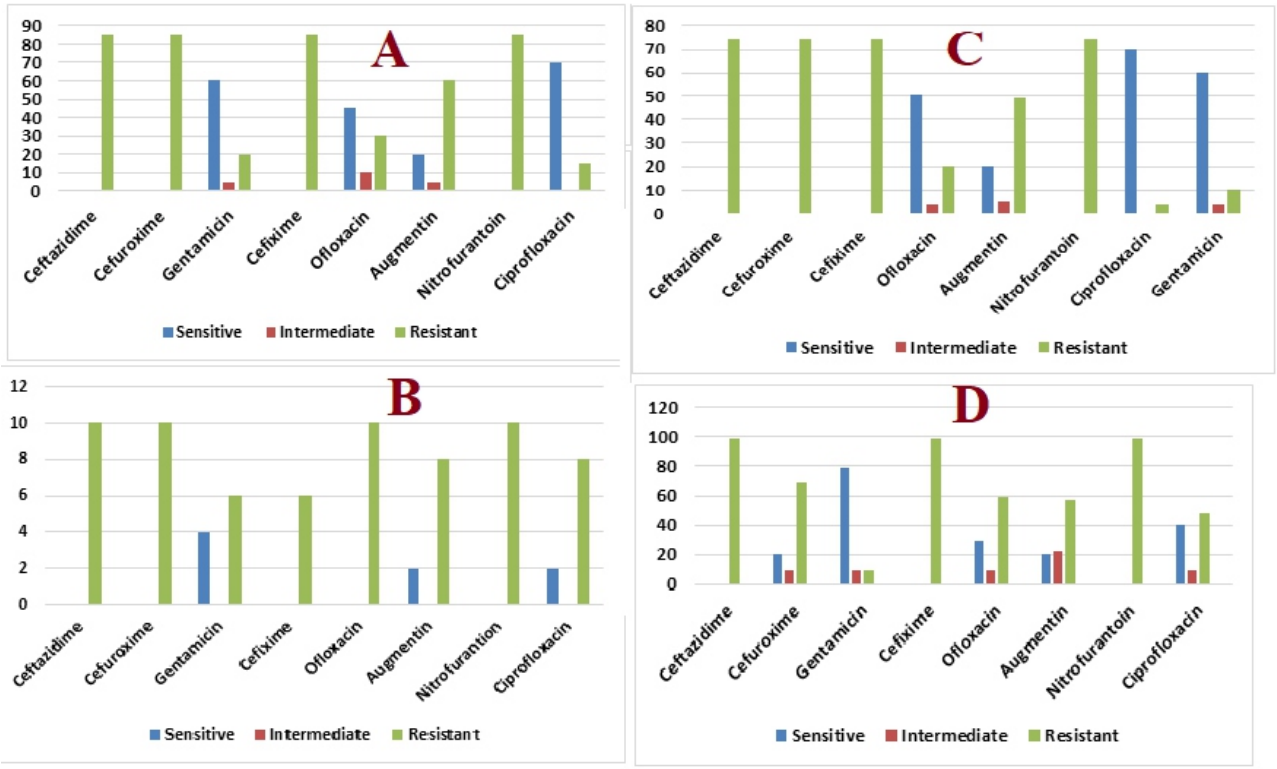

Figure 1: Antibiogram of (A*) Escherichia coli* (B) *Bacillus spp*(C)*Pseudomonas aeruginosa*(D) *Staphylococcus aureus*

4. Discussion

Currency notes, a source of legal tender for financial transaction had not been given adequate attention in many studies as a potent inanimate entity with capacity to harbor fomite and transmit pathogens of epidemiological proportion. The bank notes collected in this study were dirty, odoriferous, and mutilated and it was expected that they would not be microbe-free. Variation recorded in this study where larger percentage of feminine subjects were higher than males in some profession, could be attributed to choice of preferred profession, preference, comfort and convenience associated with various occupations. In addition to these, several studies in Nigeria have recovered from currency notes other potentially pathogenic bacteria species in similar studies including *Shigella dysenteriae, Salmonella spp, Klebsiella pneumoniae, Enterobacter spp, Aeromonas hydrophilia* and *Mycobacterium spp*[7]

Equal number of polymers to paper currency studied also varied in bacterial carriage loads, polymer notes that exists in mostly low denominations were found to carry higher microbial load irrespective of its textures, which could be due to ease of access than in higher currency denominations. The average bacterial total plate count obtained in this study were remarkable, the lowest denomination ₦5 had 4.20×10^7, ₦100 had 6.22×10^5 while ₦1000 had 2.28×10^4, which was similar to the findings of Adamu*et al,* (2012) on bacterial contaminants of Nigerian currency notes and associated risk factors, and this is suggestive of the degree of microbial contamination associated with the samples examined[8]. Microbial presence establishes the fact that these currencies could serve as a means of spreading pathogens within the populace and across different geographical boundaries.

A total of 269 bacterial samples were isolated from the currency notes examined. *Staphylococcus aureus* was found to be the highest (36.8%), followed by *Escherichia coli* (31.5%) and *Pseudomonas aeruginosa* (27.55%).

Bacillus spp were the lowest (10%) as elicited in Table 3, the variation in percentages of the isolates obtained, could be attributed to bacterial types associated with each profession and immune status of the handlers, which corroborates the study of Emikpe and Oyero[9] on In-vitro antibiotic sensitivity pattern of some bacteria isolated from Nigerian currency. Although, most of the isolates of bacterial obtained were of clinical and opportunistic status and could become pathogenic in immune-compromised subjects[9].

The antibiotic resistance pattern obtained in this study varied from one isolate to other, *Staphylococcus aureus* were 100% resistant to ceftaxidime, cefixime and nitrofurantoin, 70% to cefuroxime, 50% to ciprofloxacin but the isolates were 80% susceptible to gentamicin. The resistant patterns obtained in *Escherichia coli* also varies from one antibiotic to others, the isolates were 85% resistant to ceftaxidime, cefuroxime and cefixime while the isolates were 60% susceptible to gentamicin, 45% and 70% sensitive to ciprofloxacin. The isolates of Bacillus *spp* were 70% resistant to ceftaxidime, cefuroxime, cefixime and nitrofurantoin while the isolates were 70% sensitive to ciprofloxacin and 60% sensitive to gentamicin. *Pseudomona aeruginosa* were 70% resistant to ceftaxidime, cefuroxime, cefixime and nitrofurantoin but 60% sensitive to gentamicin and 70% sensitive to ciprofloxacin respectively. It is worrisome to note that bacteria borne on these currencies are not only pathogenic but they also showed remarkable resistance to conventional antibiotics as observed. The alarming resistance recorded as shown in figure1Figure 1A-D) in this study serves indices of contaminated currency notes as a threat to public health, which corroborates with the report of Feglo and Nkansah[10] on in-vitro antibiotic sensitivity patterns of some bacteria isolated from Nigerian currency[10].

5. Conclusion

Bacterial antibiotic resistance has been associated with treatment failure, high health cost burden and loss of manpower hours due to over hospitalization. The microbial contaminant loads capable of causing opportunistic infection were found to be present in currency notes examined. The alarming resistance of bacteria to selected conventional antibiotics used in this study, serves an indication of potential threat of contaminated currency notes to public health.

Recommendation

Public should be enlightened on the risk of microbial infections associated with abuse of currency notes, feasibility of antibiotic resistance transfer and epidemiology of treatment failures. Dirty and mutilated currency notes should not be dispensed to the public from banks and lenders hubs. Incorporation of antimicrobial substances into paper currency should be attempted to reduce the level of bacterial contaminations. Habit of using saliva to count money, keeping money on dirty surfaces, brassiere, socks, and dirty vaults should be discarded. Formalin vaporcurrency decontaminator portable currency counting machines should be cheaply available and affordable to the masses and introduction of re-washable

plastic currency notes as practiced in Australia[13] should be adopted and the use of Bitcoin and credit cards should be encouraged to limit the spread of bacteria of clinical status.

Limitations of the study

Some of the human participants contacted and briefed at the onset of the collections of currency sample notes were reluctant and skeptical about the need for our request, some demanded compensation of a little amount of money that was higher in exchange for what was collected from them but eventually succumbed to our request for sample collections.

Conflict of interest

The authors declare no conflict of interest

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