

Microbiological Evaluation of Naira Notes Handled by Fish Sellers in Umuahia Metropolis

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Abstract: Naira is the Nigerian currency used as the country's legal tender. Naira notes handled by fish sellers in Umuahia Metropolitan were evaluated for microbial contaminations. Three markets, Ubani, Orié Ugba and Ndoro were used for the study and three selling points and seven denominations of the naira were studied in each market covering 5, 10, 20, 50, 100, 200 and 500 naira denominations. The assessment involved evaluation of the microbial load, flora and occurrences of difference organisms found. $4.62(\pm 4.8) \times 10^5$ cfu/ml, Oriéugba $1.10(\pm 2.4) \times 10^5$ cfu/ml to $3.67(\pm 10.9)$ cfu/ml and Ndoro $1.27(\pm 1.7) \times 10^5$ cfu/ml to $2.49(\pm 6.8) \times 10^5$ cfu/ml). Fungal load were also recorded for the same market respectively $1.1(\pm 0.2) \times 10^3$ cfu/ml to $3.0(\pm 10^3)$ cfu/note at Ubani market, 1.8 ± 10^3 cfu/note to 3.4 ± 10^3 cfu/note at Oriéugba and Ndoro, $1.3(\pm 1.1) \times 10^3$ cfu/ml to 3.3 ± 10^3 cfu/ml. Bacterial flora showed seven isolates with percentage occurrence of *staphylococcus* 69.91%, *Escherichia coli* (52.39%), *Pseudomonas spp* (33.33%), *Bacillus spp* (66.67%), *Klebsiella spp* (42.86%), *streptococcus spp* (38.10%), Salmonella (52.39%). the occurrence of the fungi isolates were yeasts (76.19%), Aspergillus spp (52.38%), Penicillium spp (28.57%) and Rhizopus (19.05%). Statistical analysis at ($P < 0.05$) shows that there are significant differences among the different naira notes in the different markets but that there are no significant differences among the same naira notes in the three different markets. It was observed that some of the microbial isolates are known pathogens and their presence suggest the possibility of the naira note acting as the carrier of these pathogens for subsequential infection of citizens. The need for improvement in the personal hygiene of fish sellers (and other marketers) and that of the environment was highlighted and enlightenment of the marketers (buyers, sellers and others) was recommended.

Key words: Microbiological Evaluation • Naira Notes • Fish Sellers

INTRODUCTION

Money is used as a medium for exchange for goods and services, settlement of debts and for deferred payments in economic activities. Naira is the legal tender in Nigeria [1]. The naira note is a mixture of 75% cotton and 25% linen [2]. In Nigeria, the naira notes presently in circulation are abused by squeezing, stapling, sellotaping and writings on them [3]. The contamination of the naira notes could be from several sources, it could be from the atmosphere, during storage, usage, handling or

production [4]. Daily transactions have made the naira note to pass through many hands and pathogens become imposed on them before they are finally deposited in banks [5]. The contaminated currency notes go in circulation and contaminate the hands of others, transmitting microorganisms in the process [6]. Naira notes hardly last for more than one year owing to poor or negative money handling practices like spraying during ceremonies where such notes may be trampled upon when they fall on the ground [7]. Damaged, soiled or cellotaped naira notes are referred to as mutilated naira notes [8].

Microbial contaminants maybe transmitted either directly, through hand-to-hand contacts, or indirectly, via food or other inanimate objects like fomites. These routes of transmission are of great importance in the health of many populations in developing countries, where the frequency of infection is a general indication of local hygiene and environmental sanitation levels [9]. When hands used in cleaning up the anus after passing out faeces are not properly washed and are used to touch the naira note in any way, the tendency is contamination with the trophozoite of the developed parasite, eggs, cysts or even the oocyst. Other attitudes such as the wetting of hands or fingers with saliva or use of contaminated water to lubricate the hand in counting money could lead to possible transfer of parasite and bacteria from such medium to the notes [10]. Bacteria are particularly very ubiquitous and their ability to contaminate objects such as the naira notes is very prevalent when compared to parasites. Ordinarily, the exposure of naira notes to the atmosphere could even bring about contamination depending on the environment in question [11].

Paper money was introduced into Nigeria in 1945 and since then several changes has been made to different denominations we are using as legal tender. Since the note is transferable from person to person, country to country; it can act as an environmental vehicle for transmission of pathogenic microorganisms. Money is used as about 4,000 folds in each direction and lower denomination notes receives the most handling because they are exchanged many times.

The occurrence of many load of microorganisms on circulating Nigerian currency can constitute a potential health hazard to users, they carry bacteria on their surfaces and are responsible for transmitting them [12]. Dirty notes are usually moist and thus provide good surfaces for bacterial growth. They provide favourable condition such as substrate acquired from human body due to handling as well as dust from the environment. Ordinary, the exposure of money to atmosphere could even bring about contamination depending on the environment. Bank notes can be contaminated by droplets during coughing, sneezing, touching with contaminated hands and placed on dirty surfaces). Bank circulating currencies can be also contaminated by butchers and fish sellers with bloody fingers, artisans with dirty, dusty and oily fingers, teachers with chalky and inky fingers, the street vendors with wetly-oily hands etc., they will just receive or pick Nigerian bank currency notes with dirty fingers leading to contamination of the notes with microorganisms. The contaminated currencies then go in

circulation and contaminate the hand of others. Microorganisms on the skin can be transferred from cashiers, sales people and general public to the currency note that they handle.

Especially in Nigerian environment, bank currencies present a particular risk to public health hazards since the communicable diseases can spread through contact with fomites [13]. Nigeria currencies provides a large surface and breeding ground for pathogenic microorganisms, money on which microorganisms can survive represents an often overlooked reservoir for enteric diseases. Although paper currency is impregnated with disinfectants to inhibit microorganisms, pathogens are always isolated from circulating notes.

The presence of pathogenic Staphylococci on money was expected because *Staphylococcus epidermis* and disease person are common in the population. Paper money has recently been identified as mode of transmission of community-acquired *Staphylococcus aureus*. It has been suggested that human beings should keep strict adherence to hygiene practices before handling food and water after contact with bank naira notes .

Aims and Objectives:

- To isolate microorganisms associated with Nigerian naira notes used by fish sellers and identify the pathogenic ones among them.
- To characterize and identify them including the pathogenic microorganisms on Nigerian naira notes used by fish sellers.
- To investigate the level of contaminations of the Nigerian naira notes used by the fish sellers.

MATERIALS AND METHODS

Materials: All the equipment and reagents used for this project work were gotten from microbiology and biochemistry laboratories of Michael Okpara University of Agriculture Umudike and National Root Crop Research Institute Umudike, Abia state except the samples that were used for this project work which were collected from the various sampling points.

Methods

Sources of Material: The test naira notes were obtained from randomly selected fish sellers in three markets covering Ubani, One Ugba and Ngoro markets. Laboratory and other facilities used in the practical work

were sourced from central laboratory services unit of National Root Crops Research Institute, Umudike in Association with the microbiology laboratory of Michael Okpara University of Agriculture Umudike.

Sample Collection: Sample of Naira notes used in the project work were collected aseptically. Hard gloves were warm and fresh ones were used for each point of collection. Only the denomination below N1000 were used and fish sellers were informed of the purpose and currency note were exchanged in return for equal value and in some cases fish was bought and the change collected. The collected currency notes were put in a sterile polythene containers and taken to the laboratory initiating after collection.

Media Preparation: The media used, nutrient Agar and in summarized Dextrose Agar were prepared in accordance with the manufacturing directives. Exactly 28g of NA and 65g of SDA and 63g of SSA were separately dissolved in 1 liter of distilled water in flasks. The flasks were heated in a water bath until the agar in each case melted. The SSA was sampling heated on a Clifton heater until it boiled while the smear media were sterilized by autoclaving at 121°C for 15 minutes. They were each allowed to cool to about 45°C before they were aseptically poured in 15 ml portions into petri dishes where they were allowed to cool. The sterile solid plates were used for the microbial culture.

Isolation of Microorganisms: The method described by [3] was used. Each test currency was aseptically (with the aid of a flamed pair of forceps), was placed on a surface, sterilized bench top. Carefully, wet sterile swab sticks was used to roll over the surface of the note and then placed in a test tube containing 10mls of sterile distilled water. A sterile forcep was used to push out the cotton wool of the swab stick into the water in the test tube. The tube was corked and shaken vigorously (to dislodge the microbes from the swab wool. Thereafter the swab wool removed after pressing it by the side of the tube to squeeze out the water. The water in the test tube (containing the dislodged microbes), was distilled serially to the fourth diluent 10^{-4} . From the 2nd diluent (10^{-2}) inoculate of 0.1ml each were collected and aseptically inoculated into the surface of solid potato Dextrose agar. With the aid of a flamed glass hockey, the inoculum, was spread over the surface of the medium in plate (Spread plate technique). The same way, inoculum of 0.1ml was taken from the 4th diluent (10^{-4}) and plated by spread plate technique on Nutrient Agar, MacConkey Agar and

Salmonella shigella agar separately. Each inoculation was due in triplicates. The PDA plate (for fungi) was incubated at room temperature (28 - 32°C) for 2 - 5 days while the bacteria culture plates [NA, MA and SDA] were incubated at 37°C for 24 to 48 hours. The plates were observed daily for growth. On establishment of growth, the number of colonies in each plate was counted by means of a digital colony counter. The microbial load was calculated as the total viable count expressed in colony forming units per currency.

Isolation of Microbes: Isolation of microorganisms (fungi and bacteria) was done by sub-culture from the initial sample culture plates. Following the establishment by growth, each sample culture plate was examined for distinct other colonies. From such colonies, inoculate were collected and transformed aseptically onto fresh sterile medium in plate. After incubation, the plates were examined for uniformity as make of purity. The pure cultures were used for characterization and subsequent identification.

Characterization of Fungal Isolates: Fungal isolates from the tests Naira notes were characterized based on their cultural and structural characteristics. Each pure culture plate was examined closely virally and characteristic features of the colonies were recorded including, extent of growth, color, pressure of mycelia, spore etc. Furthermore slides mount of the isolates were made stained with lactophenol cotton blue stain. Structures like sporangiophore, conidiophore etc were observed and their form, duration of growth, septation, branching etc were recorded as well as the color and relative size of spores, conidia etc were also recorded and were compared with a mycological atlas .

Characterization of Bacteria: Bacteria isolates were characterized in a four step approach as follows:

Colony Features: The colonies were observed and their respective features were recorded including extent of growth, color, elevation, pigmentation form and edge of colonies, consistency etc.

Microscopic Features: Structural features and microscopic characteristic were observed under the microscopes and recorded including; reaction to several dye (using Gram stain), reaction to specific dyes to show the presence of features like capsule flagella etc. also the cell shape and arrangement as well as motility test were also recorded.

Biochemical Tests: This involved tests to show the ability of the isolates to produce enzymes which include catalase. Oxidase, urease and coagulase. Other tests include ability to reduce nitrogen, utilize citrate etc. in addition to methyl red, indole and voges proskeur tests.

Sugar Utilization Test: In this case, different sugars like glucose, lactose, sucrose, maltose and manitol xylose etc. were separately incorporated into the broth media containing indicator and inverted duham tube. Each isolate was cultured in those media and after incubation was kept on their ability to utilize the sugars as shown by change in color (acid production) and production of gas (shown by trapped gas in the inverted duham tube).

Identification of Isolate: Identification of isolates was based on matching their respective characteristics with those available in standard manuals. Fungi isolates were matched against available taxa illustrated Genera of important fungi while bacteria isolates were matched with standards in Berge's manual of determinative bacteriology.

Determination of Occurrence: Occurrence of each isolate was calculated as a percentage ratio of their respective produce

Statistical Analysis: The statistical analysis were carried out using SPSS and ANOVA according to [11].

RESULTS

Result gotten from the analysis of data collected according to the different naira denomination and locations as shown in Table 1 with ₦500 notes at ubani having the highest bacterial load and ₦10 notes at ubani also having the lowest bacterial load.

Table 2 shows the analysis of data carried out for fungi with ₦200 and ₦500 notes showing the highest fungi load at Oriegba Market and ₦5 notes having the lowest fungi load at ubani. Other tables showing results on the morphological characteristics of fungal and bacterial isolates in Table 3 and 4 respectively. In Table 5 showing results on the biochemical tests carried out during the projects work. Figure 1,2,3,4,5 and 6 showed the percentage occurrences of bacterial and fungal isolates in the different denominations in naira notes in the 3 different markets Ubani, Oriegba and Ndoro.

Table 1: Data analysis showing bacteria load occurrence of naira note (x10⁴cfu/note)

Notes (₦)	Ubani(x10 ⁵)	Oriegba (x10 ⁵)	Ndoro(x10 ⁵)
5	1.08(± 2)	1.10(± 2.4)	1.24 (± 1.7)
10	8.0 (± 1.2)	1.01(± 4.2)	1.29(± 3.2)
20	1.01 (± 1.9)	1.27 (± 3.3)	1.50 (± 2.9)
50	1.26 (± 7.2)	1.51 (± 7.7)	1.48(± 5.6)
100	1.73 (± 1.2)	2.35(± 7.3)	2.10(± 3.1)
200	2.59 (± 7.3)	2.91 (± 9.5)	2.17 (± 7.0)
500	4.62 (± 4.8)	3.67 (± 10.9)	2.49 (± 6.8)

Table 2: Fungi Load Occurrence Of naira Note (x10³ cfu/note)

Notes (₦)	Ubani (x10 ³)	Oriegba (x10 ³)	Ndoro (x10 ³)
5	1.6 ± 0.4	1.8 ± 0.4	1.7 ± 0.4
10	1.3 ± 0.0	2.1 ± 1.1	1.3 ± 0.4
20	2.7 ± 1.7	2.1 ± 0.7	2.1 ± 0.3
50	1.1 ± 0.2	1.6 ± 0.2	2.3 ± 0.9
100	3.0 ± 0.6	1.8 ± 0.4	2.1 ± 0.5
200	2.8 ± 0.5	1.9 ± 1.0	3.0 ± 0.7
500	3.1 ± 1.5	3.4 ± 0.8	3.3 ± 0.9
Gross Mean	2.2 x 10 ³	2.1 x 10 ³	2.3 x 10 ³

Values show means of triplicate analysis ± Standard deviation. Figures with different superscript down the column are significantly different (P < 0.05).

DISCUSSION

Results from laboratory analysis of the test Naira currency notes are shown in table 1. Table 1 shows that bacteria loads of the different currency notes obtained from fish sellers in the three study areas:of Ubani, Oriegba and Ndoro. The result shows a significant difference (P < 0.05) in the bacteria load of the different currency notes. In Ubani, fish sellers currency notes, ₦500 notes haboured the highest population of bacteria with a mean load of 46.2 ± 4.8 x 10⁴ colony forming units per note while ₦10 notes had the least load of 8.0 ± 1.2 x 10⁴Cfu/note. Similar variations of significant difference were recorded in Oriegba and Ndoro fish sellers currency notes. In Oriegba, the bacteria load ranged from 11.0 ± 2.4 x 10⁴ to 36.7 ± 10.9 cfu/Note while the range at Ndoro. The bacteria load of the currencies ranged between 12.7 ± 1.7 x 10⁴cfu/ml to 2.49 (± 6.8) x 10⁵cfu/ml. In all the test areas, smaller non-paper currency notes of ₦5, ₦10, ₦20 and ₦50 contain relatively lower bacteria load than the higher paper notes of ₦100, ₦200, ₦500 notes. Comparatively, the fish sellers currency at Ndoro market had the least amount of bacteria load (1.76 x 10⁵ cfu/ml mean) while those at Oriegba had the highest bacteria load of 1.97 x 10⁵ cfu/ml as mean load.

Table 2 shows the average fungal load of fish sellers Naira notes in the different markets. In Ubani market, the average fungal load was between 1.1(± 0.2) x 10³ cfu/ml

Table 3: Showing Morphological characteristics of fungi isolates

Biochemical	Isolate	Isolate	Isolate	Isolate
Characters	1	2	3J	4
Colony	Black colonies	Brownish colonies	Blue, green colonies	
Morphological	Septate branched,	Cottony, surface, it is	Septate	
Characters	Mycelium, Blackish conidia, Ascospores as Produced	Non-septate and forms Chlamydo pores	Branched, Mycelium, With Conidiophores	
Possible fungus	<i>Asperillus Spp</i>	<i>Rhizopus spp</i>	<i>Pencillin spp</i>	<i>Yeast</i>

Table 4: Showing Morphological characteristics of bacterial isolates

Characters	Isolates							
	1	2	3	4	5	6	7	8
1. Pigment characters			Yellow		Golden yellow to orange		Smote round florescent greenish	
2. Motility	Motile	Motile	Motile					
3. Gram Reaction	Gram negative	Gram negative	Gram positive	Gram positive	Gram Positive		Gram negative	Gram negative
4. Spore formation	Negative	Negative	Negative	Positive	Negative		Negative	Negative
5. Shape	Straight rods <i>Salmonel la spp</i>	Rods <i>E. coli spp</i>	Cocci in tetrads. Pairs <i>Streptococcus spp</i>	Rod <i>Bacillus spp</i>	Cocci <i>Staphylococcus spp</i>		Rods shorts chains <i>Pseudomonas spp</i>	Rod shape <i>Klebsiella spp</i>

Table 5: Showing biochemical characteristics of bacterial isolates

Biochemical	Isolate	Isolate	Isolate	Isolate	Isolate	Isolat	Isolate
Characters	1	2	3	4	5	e	7
						6	
1. Catalase	Positive	Positive	Negative	Positive	Positive		
2. Indole	Negative	Negative	Negative		Negative		
Production							
3. Methyl red	Positive	Positive	Positive		Positive		
4. Voges - Prosakauer	Negative	Negative	Negative		Positive		
5. Coagulase	Negative	Negative	Positive		Positive		
6. Glucose Utilization	Positive	Positive	Positive		Positive		
7. Citrate	Positive	Negative	Negative		Positive		
	<i>Salmonella</i>	<i>E. coil</i>	<i>Streptococcus</i>	<i>Bacillus</i>	<i>Staphylococcus</i>	<i>Klebsi ell a sp</i>	<i>Pseudom onas sp</i>

and $3.0(\pm 0.6) \times 10^3$ cfu/ml. The ranges in the other markets are Oriuegba $1.8 (\pm 0.4) \times 10^3$ cfu/ml to $3.4 (\pm 0.8) \times 10^3$ cfu/ml and Ndoro, $1.3 (\pm 0.4) \times 10^3$ cfu/ml to $3.3 (\pm 0.9) \times 10^3$ cfu/ml. This shows that some Naira notes of fish sellers in Ndoro had the highest load of fungi with a gross mean load of 2.3×10^3 c cfu/ml whereas those of sellers in Oriuegba had the least 2.2×10^3 cfu/ml.

The high microbial load of the currency notes of fish sellers was perhaps due to two reasons. First, the neighbourhood environment in the market were not of high level sanitation, in addition to the questionable personal hygiene of the fish sellers. Secondly, The nature of the fish being sold almost always presents a moist hand which moistens the note as it changes hand, thus, preparing a good ground for the germination of microbial spores.

The bacteria flora and their occurrences in the different denominations of the fish sellers' Naira notes are shown in Figure 1, 2, 3 and Figure 4, 5, 6. The specie of bacteria isolated in the various denominations of the naira notes in the different markets include *Staphylococcus spp*, *E.Coli*, *salmonella spp*, *pseudomonas spp*, *Bacillus spp*, *kliebsella spp* and *streptococcus spp* but at varying occurrences in the different notes. The results show variations in the occurrence of bacteria in the different Naira notes. In Ubani market, more bacteria species were seen more in the □200 denominations (90.5%) than the rest. The □5 notes in all the markets haboured the least number of bacteria species with percentage occurrences of 19.4%, 14.29% and 33.29% at Ndoro, Oriuegba and Ubani markets respectively.

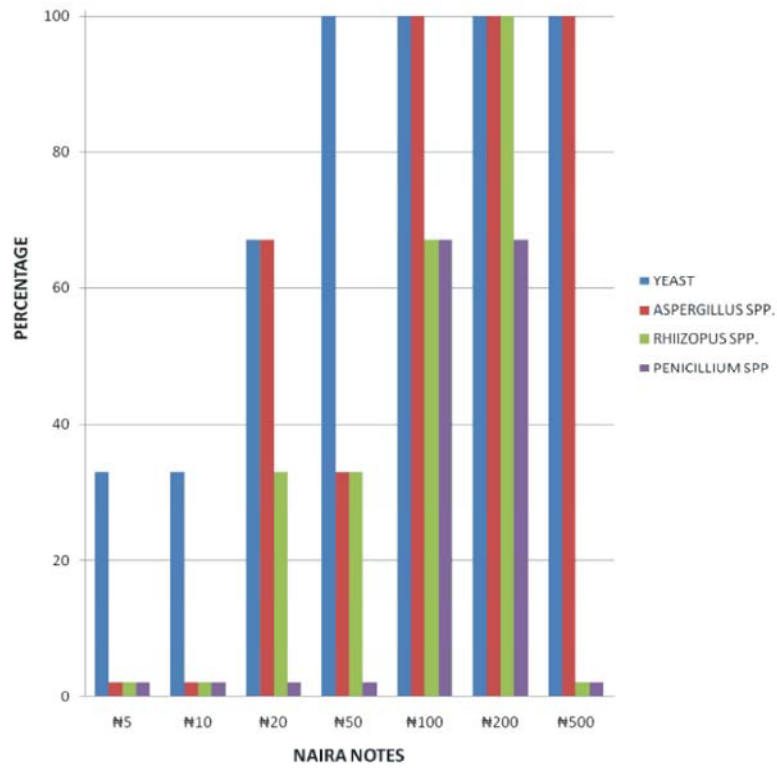


Fig. 1: Percentage Occurrence of Fungal from Naira Notes of Fish Sellers at Orié Ugba Market

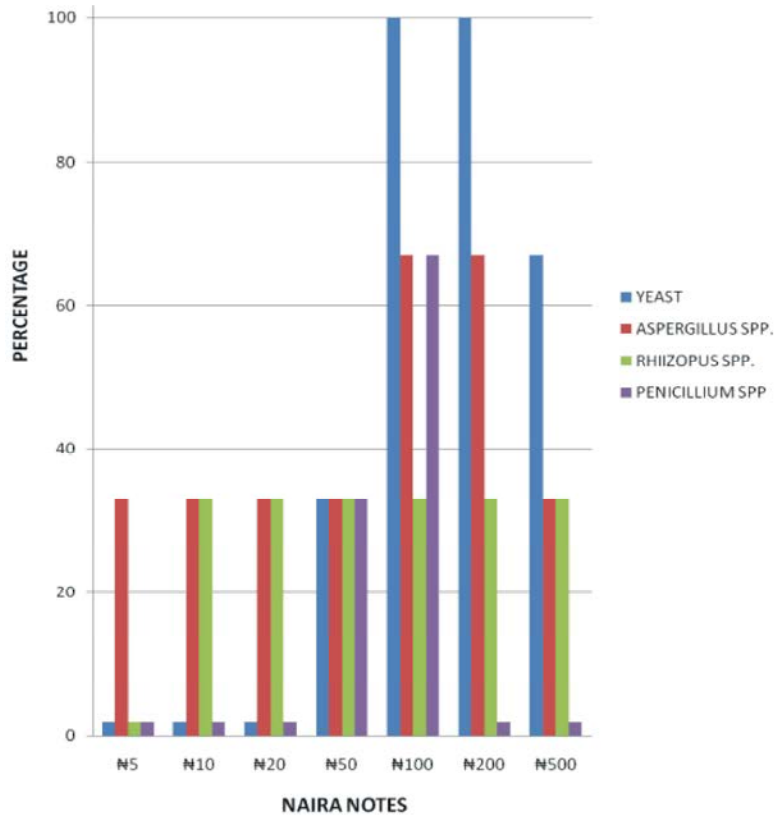


Fig. 2: Percentage Occurrence of Fungal isolates from Naira Notes of fish sellers at Ubani Market

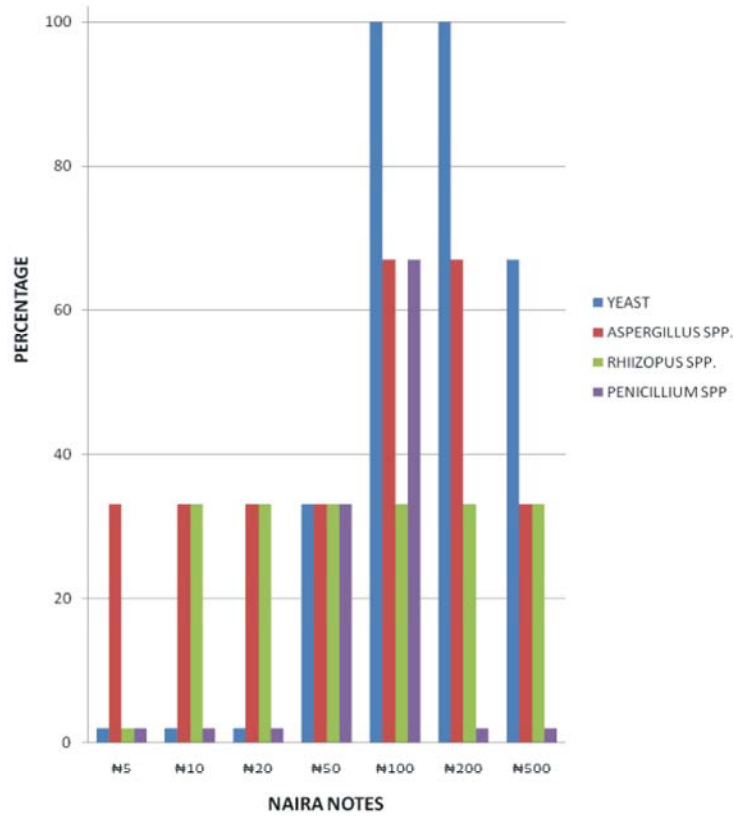


Fig. 3: Percentage Occurrence of Fungal Isolates from Naira Notes of Fish Sellers at Ngoro Market

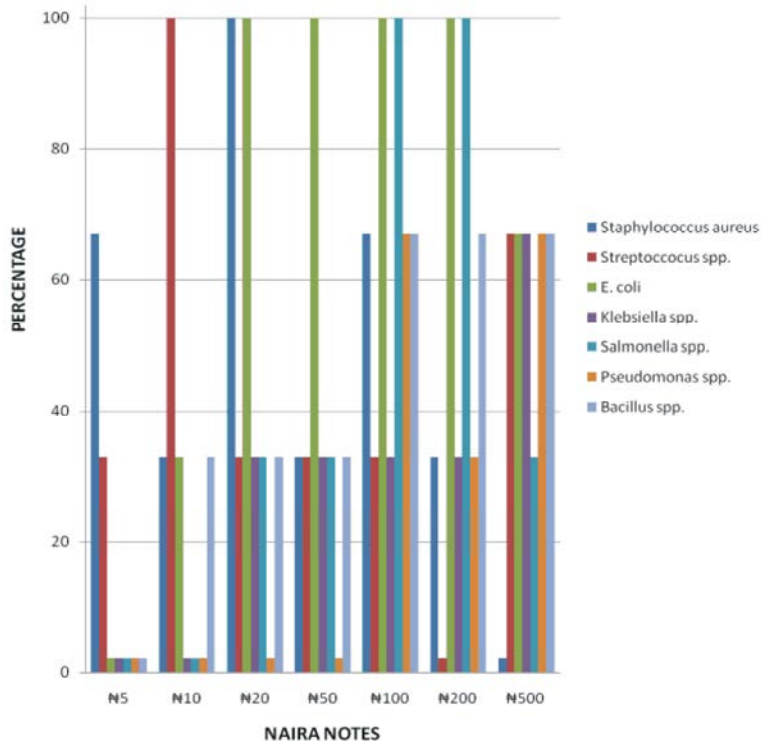


Fig. 4: Percentage Occurrence of Bacterial Isolates from Naira Notes of Fish Sellers at Oriu Ugba Market

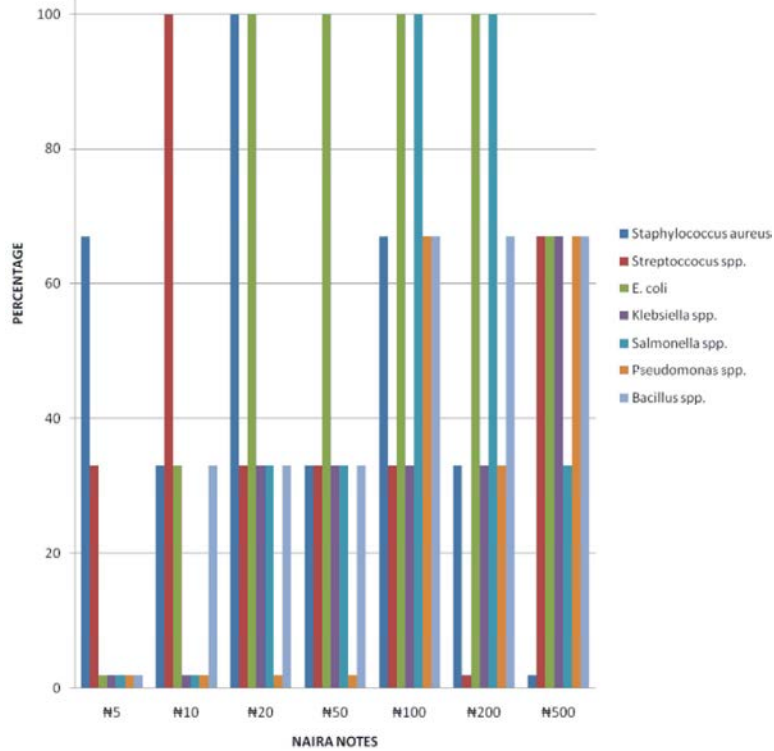


Fig. 5: Percentage Occurrence of Bacterial Isolates from Naira Notes of Fish Sellers at Ubani Market

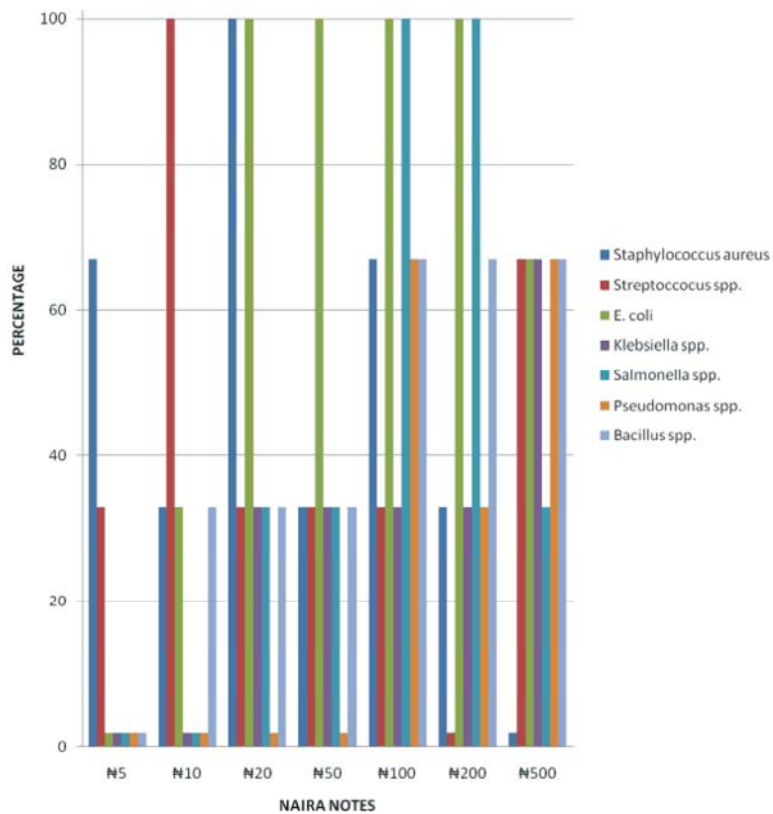


Fig. 6: Percentage Occurrence of Bacterial Isolates from Naira Notes of Fish Sellers at Ndoro Market

At both Ndoro and Oriugba markets, the highest occurrence of the different bacteria species in the ₦100 denomination with occurrence of 94.43% and 66.71% respectively. In comparison, less of the different bacteria species were found in the notes at Oriugba (14.29% - 66.71%) than in Ndoro (19.14 – 95.43%) and Ubani (33.29 – 90.57%). Fungi species isolated and identified from the different currency notes include species of *rhizopus spp*, *penicillium spp*, *aspergillus spp* and yeasts but with varying levels of occurrences in the different notes. Similar varieties were also recorded in the occurrences of the different fungal species. The range was 8.25% in ₦5 to 91.75% in ₦200 note in Oriugba. In Ubani it was 8.25% in 5 note to 91.75% in 100 note. while the range was 8.25% to 66.75% in ₦100 note at Ndoro market. Generally, there were less of the species of fungi in the poly notes ₦(5, 10, 20 and 50) notes than the paper notes of ₦(100, 200, 500).

The different species of microorganisms isolated from the samples were found to confirm previous research findings. In Bangladesh, isolated similar bacteria and fungi from 'Taka' the paper currency of the country. The issue of microbial contamination of currency notes has been reported from different parts of the world [10, 11 and 12].

The occurrence of staphylococcus in most of the currency notes was considered to be perhaps due to rub off from humans; as the organism is a normal flora of the human body [6]. The presence of different species of bacteria and fungi in the currency notes (Naira). Confirms potentials of ability to transmit diseases as some of the microbes are known to be pathogenic.

CONCLUSION

From findings in this project work, it was concluded that a wide range of different species of bacteria and fungi contaminate the naira notes used by the fish sellers in the different markets studied. Variation of significant difference ($P < 0.05$) were recorded at both the load and the flora of the different Naira denominations. Findings also show that the very low denominations ₦(5, 10, 20 and 50) were less contaminated than the higher denominations of ₦100 to ₦500. Also, the work reveals that the level of microbial contaminations vary with the markets and the currency notes. The species of microbes isolated from the different notes in the different markets show clearly that pathogens are harboured by the notes and this was considered a threat to the safety of the money users.

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