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Original Research Article

MICROBIOLOGICAL EXAMINATION OF HOUSEHOLD KITCHEN SPONGES FROM THREE COMMUNITIES IN IKWUANO L. G. A, UMUAHIA, ABIA STATE NIGERIA.

ABSTRACT

Sixty kitchen sponges collected from the households in three major communities in Ikwuano L.G.A, Umuariaga, Amawom and Amaoba were examined microbiologically using standard methods. *Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa* and *Aspergillus* niger were recovered from the sponges. The Total Heterotrophic Plate Count (THPC) for Umuariaga community ranged from 4.0 x 10^8 – 1.73 x 10^9 cfu/ml with a mean of 8.02 x 10^8cfu/ml ,_3.65 x 10^8 -1.86 x 10^9cfu/ml with a mean of 9.13 x 10^8 cfu/ml and 4.65 x 10^8 - 1.93 x 10^9 cfu/ml and Total Coliform Plate Count (TCPC) that ranged from 3.60 x10^7 - 9.7 x10^8 cfu/ml with a mean of 6.11 x10^7 cfu/ml. Amaoba community has THPC with a mean of 9.13 x 10^8 cfu/ml and with a range of 3.65 x 10^8 - 1.86 x 10^9 cfu/ml while the mean TCPC was 6.70 x 10^7 cfu/ml with a range count of 3.59 x 10^7 - 1.06 x 10^8 cfu/ml. The mean THPC for Amawom community was 8.47 x 10^8 cfu/ml with a range count of 4.65 x 10^8 - 1.93 x 10^9 cfu/ml while the mean TCPC was 6.28 x 10^7 with count range of 3.55 x 10^7 - 1.01 x 10^8 cfu/ml. Antibiotics susceptibility pattern of the isolates showed that *Staphylococcus aureus* was most sensitive to Ofloxacin (21.4mm) but resistant to Gentamycin and Nalidixic acid while *Escherichia coli* was most sensitive to Amoxicillin (22.0mm). *Pseudomonas aeruginosa* showed the highest sensitivity to Ofloxacin (20.6mm) and incidentally the highest resistance (to five antibiotics). Based on the results, it was evident that kitchen sponges can be contaminated by pathogenic microorganisms which showed varied levels of susceptibility to antibiotics. Regular change of kitchen sponges and the use of washing disinfectants can reduce the level of contamination of the kitchen sponges and the associated infections.

Key words: Bacteria, coliform, contamination, fungi, infections, kitchen sponges.

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1.0 INTRODUCTION

It is known that during the cleaning process of equipments utensils, sinks in the kitchen, the pre-washing and washing steps are done with the use of sponges to eliminate food residues. As a consequence of these procedures, part of the residues adheres to the kitchen sponges surfaces. These food residues together with the moisture retained in the sponges offers a favourable environment for bacterial growth, and this may lead to the formation of biofilms [1].

The kitchen sponges are made of materials such as fiber and cellulose [2], this aid in harbouring microorganisms that are capable of causing serious infections thereby posing a constant risk of transferring contaminants. Kitchen sponges have been recognized as important diffusers of pathogens and they cause cross contamination in food leading to foodborne diseases. These sponges can serve as a reservoir of foodborne pathogens. Contaminated sponges can transfer pathogens to surfaces that come in contact with foods, and these microorganisms can remain viable on these surfaces for hours or days after contamination [3].

There has been a significant increase in foodborne illnesses worldwide. The microbial causes of foodborne illnesses include bacteria such as *Escherichia coli, Staphylococcus aureus,* Enterobacteriacea*, Pseudomonas* spp*, Salmonella* spp*,* Parasites such as *Entamoeba histolytica, Trichinella spiralis* and some viruses with symptoms ranging from mild gastroenteritis to life threatening neurologic, hepatic and renal diseases. Diarrhoeal diseases alone, kills a considerable proportion of 2.2 million people globally [4]. These foodborne diseases can result from cross contamination from the household kitchen to other materials that come in contact with food such as kitchen sponges.

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Many household kitchen sponges are kept at room temperature, inside containers with water and food residues which can contribute to microorganism multiplication. Effective hygienic procedures are important in the prevention of cross contamination, bacterial growth and survival in the kitchen sponges. Disinfection of sponges may prevent the survival and spread of pathogens in the kitchen. As life style changes, individuals spend more time on the kitchen preparing meals and subsequently give less attention to proper food handling and sanitation practices that can reduce foodborne diseases and spoilage of food [5]. Simple fast and effective methods to disinfect kitchen sponges may prevent the spread of pathogenic microorganisms in household kitchens and may lead to better food preservation and fewer cases of foodborne illnesses [6]. To this end, this research work which was carried out in 2015 aimed at microbiological examination of sampled kitchen sponges in three communities of Ikwuano L. G. A and determining the susceptibility patterns of the isolates.

2.0 MATERIALS AND METHODS

2.1 STUDY AREA

The kitchen sponges in this research work were collected from three different communities in Ikwuano Local Government Area of Abia state namely: Umuariaga, Amawom and Amaoba. The major population of this area is dominated by peasant farmers and low income workers.

2.2 SAMPLE COLLECTION

Random households were selected in each community and kitchen sponges were collected by going door to door requesting for one's used sponges from the kitchen. The sponges sampled were in use for at least 2 weeks and this information was confirmed by the person who provided the sponge. The sponges were collected with latex gloves and placed in sterile plastic bags with

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appropriate labels being stored in a cooler containing ice pack at a temperature of 5°C. The sponge was transported to the laboratory 4 hours of collection and was analyzed within 24 hours. A total of 60 sponges were collected. The method of sample collection described by [7] was used.

2.3 SAMPLING PROCESSING AND ANALYSES

Each kitchen sponge was weighed and 75-100ml of peptone broth was added to the sponges placed inside a sterile 500ml beaker to guarantee full soaking of the sponge. This sponge was manually kneaded in the peptone broth for five minutes until the broth was completely absorbed by the sponge. The broth was extracted from the sponge by wringing the liquid out aseptically [8]. 1ml of the extracted broth was serially diluted in peptone water and 0.1ml aliquot of appropriate dilution was inoculated onto Nutrient, MaConkey and Sabourand Dextrose agar respectively. The Nutrient Agar plates (Titain Biotech, limited) were used to determine the Total Heterotrophic Plate Count (THPC) while the MaConkey agar plates (Titain Biotech, limited) were used to determine the Total Coliform Plate Count (TCPC). Sabourand Dextrose agar (Titain Biotech, limited) containing 0.1g/l chloramphenicol to inhibit bacterial contaminants was used for determining the Total fungal count. All the inoculated petri dishes were incubated for 37°C for 24 hours for the isolation of bacteria while the SDA was incubated at 25°C for the isolation of fungi. The plates were incubated prepared in duplicates while uninoculated plates were kept as control. After incubation, plates containing 30-300 colonies were chosen for enumeration. The results were expressed in cfu/ml [8].

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2.4 IDENTIFICATION AND CHARACTERIZATION OF ISOLATES

Cultural and microscopic examinations were carried out. Colonies were selected using their morphological characteristics (sizes, pigmentation, elevation, consistency). Gram staining, lactophenol cotton blue staining, biochemical and sugar fermentation tests were performed [9].

2.4.1 LACTOPHENOL COTTON BLUE STAINING

A drop of 70% alcohol was placed on a microscope slide and the fungal isolate was immersed on the drop of alcohol. Two drops of the lactophenol cotton blue mountant was added before the alcohol dried out. With the cover slip held between the fore finger and thumb, one edge of the mountant was touched and lowered gently avoiding air bubble and then viewed under the microscope [10].

2.5 ANTIBIOTIC SENSITIVITY TESTING

The sensitivity pattern of the pathogenic isolates was determined using the Kirby-Bauer disc diffusion technique [9]. The 24 hour broth culture of the isolates was matched to the turbidity of 0.5 McFarland standards in 0.9% saline and then streaked uniformly on Mueller-Hinton agar plates using swab sticks and allowed to stay for 15minutes. Antibiotic disc was then aseptically placed at reasonable equidistance on the inoculated Mueller-Hinton agar plates using sterile forceps and then incubated at 37°C for 18 hours. The diameter of the zones of inhibition produced by each antibiotic disc was measured using transparent 15cm rule and recorded in milimetre.

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3.0 RESULT AND DISCUSSION

The microbial isolated recovered from the 60 analyzed kitchen sponges were *Escherichia coli, Staphylococcus aureus, Pseudomonas aeruginosa* and *Aspergillus niger* (Tables 1 and 2). The distribution of the microbial groups shows that *Escherichia coli* had 100% occurrence in all the analysed kitchen sponges followed by *Pseudomonas aeruginosa, Aspergillus niger* and *Staphylococcus aureus* with 43.3%, 17.3% and 13.3% occurrences respectively.(Table 3)

The presence of *E. coli* (a faecal indicator organism) is an indication of faecal contamination. This could present a danger to health, especially for populations at risk including the very young, the elderly and immune-compromised. *E. coli* is responsible for infant diarrhoea and gastroenteritis and its presence in all analyzed kitchen sponges could be due to contaminated water. This would promote cross contamination to the kitchen sponges and the cooking utensils washed with the contaminated water. [11] isolated 54.1% of *E. coli* in kitchen sponges used in the United States and this is in agreement with the findings here.

Pseudomonas aeruginosa which has a percentage occurrence of 43.3%. This bacterium is an opportunistic pathogen responsible for many hospital acquired (nosocomial infections). It thrives in many atmospheric conditions and colonizes natural and artificial environments and thus being able to colonize kitchen environments. Several studies have shown that dish cloths, sponges and towels used daily in the kitchen contribute to the transmission of pathogenic microorganisms. A study of dish cloths showed that 37% of them were contaminated with *L. monocytogenes* and 18 of the 108 analyzed cloths were contaminated with this bacterium at levels ranging from 1000 to 10,000 CFU per cloth [12]. [13] studied the relationship between bacterial contamination of countertops and cleaning cloths. When used for cleaning in a professional caterer's kitchen

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during working hours, detergent-soaked cloths were heavily contaminated within a few hours. After cleaning of the premises and equipment, their level of contamination and the contamination of the cleaned surfaces were found to be even higher, which indicates the transfer of bacteria from the cloths to the surfaces and vice versa. [14], in a study on cleaning cloth contamination, confirmed that while cloths could play an important role in the transfer of bacteria to surfaces, there was no significant difference between the microbial load of wet cloths, dry cloths, those used for short and prolonged periods and cloths used for different activities.

kitchen sponges are used through direct contact with hands during washing,therefore, presence of *S. aureus* on the sponges could be due to transfer from human hands were this pathogen live naturally [15]. However, only 13.3% of sponges had *S. aureus* in them and this could be due to the antimicrobial effect of the washing soaps and detergents used by the homes. *S. aureus* causes staphylococcal food poisoning and may also be present in foods such as meat, poultry and egg products. Gram positive bacteria such as *S. aureus* have a cell wall which is more sensitive to anionic surfactant present in some detergents which can contribute to the inactivation of these microorganisms. Other facts that can explain the low percentage of *S. aureus* in sponges is could be due to high water activity in the sponges and inter-microbial competition in the sponges [11]. Lower results have been found by [16] who isolated 2.7% of *S. aureus* in kitchen sponges used in Netherlands.

Sponges are used frequently to clean surfaces. [17], in a survey of French consumers showed that 89% of respondents said they used a sponge to clean their refrigeratosr. Due to their large surface/volume ratio, their almost constant humidity and the nutrients for bacterial growth they can contain, sponges are an ideal habitat for bacteria [18]. [16] studied the microbial quality of sponges used in domestic kitchens in Mexico. They showed that 9.8% of them were

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contaminated with Salmonella and that 60% of cellulose sponges and 86% of natural sponges were contaminated with *S. aureus*. In contrast, in a North American study published the same year, only 4% of the sponges were contaminated by *S. aureus* and none by Salmonella or Campylobacter [14].

The only fungal species isolated in the analyzed sponges was *Aspergillus niger* with 17.3% occurrence (Tables 2 and 3). This pathogen is present in some type of foods especially grains and cereals where it produces some toxins which are life threatening. Damp environment favours its growth and this could be one of the reasons for its survival in kitchen sponges. *A. niger* causes some types of infections in humans such as aflatoxicosis.

The bacterial load of the kitchen sponges was high and varied between the different locations. From Tables 4, 5 and 6, the sponges had Total heterotrophic plate counts (THPC) that ranged from 4.0×10^8 -1.73×10^9cfu/ml with a mean of 8.02×10^8cfu/ml; 3.65×10^8 -1.86×10^9 cfu/ml with a mean of 9.13×10^8 and 4.65×10^8 – 1.93×10^9 cfu/ml with a mean of 8.47×10^8 for the three locations namely Umuariaga, Amaoba and Amawom respectively. This study shows that sponges can be contaminated with a diverse group of organisms, supporting the results found by other researchers. For example, heterotrophic microorganism contamination was also found by [3] who observed approximately 6 log cfu/ml of hetetrophic microorganism in kitchen sponges used in Netherlands. [19] found 6.9 log cfu/ml of heterotrophic microorganisms in kitchen sponges used in Turkey.

Total coliform plate count (TCPC) from the kitchen sponges ranges from $3.60 \times 10^7 \times 9.7 \times 10^7$cfu/ml with a mean of 6.11×10^7 cfu/ml; 3.59×10^7 - 1.06×10^8 cfu/ml with a mean of 6.70×10^7 and 3.55×10^7 - 1.01×10^8 cfu/ml with a mean of 6.28×10^7 cfu/ml for the three locations namely Umuariaga, Amaoba and Amawom respectively (Tables 4, 5 and 6). Presence of

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coliforms in kitchen sponges indicates the presence of faecal materials and may indicate the presence of foodborne pathogens. Coliform contamination in sponges can be attributed to different factors such as inappropriate hygienic and sanitary practices during food preparation. When the average counts of coliforms where compared from the various locations, the highest number of contamination by coliforms were obtained from sponges collected from Amawom, this could be attributed to the low hygienic standard of the rural community.

The higher number of kitchen sponges contaminated by various microorganism in the present research which was conducted in rural communities dominated mainly by population of middle to low income residents when compared to the results found by other researchers who conducted their research mainly in developed areas dominated mainly by high income workers may be attributed to factors such as lack of use of self-disinfecting sponges which are used in developed countries. Self-disinfecting sponges which is often colonized by lower numbers of microorganism in comparison to regular sponges reduces the transfer of total and faecal coliform bacteria to surfaces and to hands [20].

Inadequate disinfection methods contributed to higher number of microbial contamination of our kitchen sponges. Sanitizers should be also used so long as they are used in accordance with the manufacturer's use direction included in the labeling. The aim is to sanitize the sponges after they have been contaminated through use. Sanitization is the cumulative effect of treatments that results in at least 5-$\log_{10}$ (99.999%) reduction in representative disease microorganism of public heath importance [21].

The method of washing in the kitchens of developed areas makes use of running tap waters which has greater flow rate and thus displaces entrapped microorganisms. The rural communities

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make use of bowl water which has low flow rate and thus microorganisms are not easily removed leading to the formation of biofilms. Most of the sponges in the rural communities are stored in a moisture laden environment which promotes the fast colonization and multiplication of microorganisms as opposed to developed areas where sponges are kept in air dry condition. Delay in the washing of cooking utensils leads to the rapid multiplication of microorganism which could be cross-contaminated to the kitchen sponges during washing. Most sponges used in the rural communities are made of fibre and there are also natural sponges this also leads to rapid colonization as opposed to those in developed areas which are made of cellulose and cottons. There are usually enough nutrients remaining in dish cloths and sponges to support the growth of most bacteria. If the sponges and dishcloths are dried after use, the bacterial growth is halted. However, the best practice is to use a clean sponge or dish cloth each day and to use a brush for washing dishes and kitchen utensils. The brush can be rinsed out easily and dried rapidly. Among other answers to this problem is the FDA Food Code recommendation that cleaning cloths should be kept in the sanitizer bucket. But this practice does not prevent the microbial population from increasing because after sometime, the organic material obtained from cleaning neutralizes the sanitizer and the microorganisms begin to multiply [21].

Antibiotic sensitivity testing (Table 7) of the isolates showed that all the bacterial isolates were susceptible to Ofloxacin which also showed highest sensitivity for *S. aureus* and *P. aeruginosa* (21.4 and 20.6mm respectively). However, *S. aureus* was resistant to Amoxicillin (14.0mm), Gentamycin (7mm) and Nalidixic acid (10.0mm) respectively. *Escherichia coli* was most sensitive to Augmentin (23.5mm) but resistant to Nitrofurantoin and Gentamycin. *P. aeruginosa* showed the highest resistance among the bacterial isolates being resistant to Amoxicillin (8.5mm), Augmentin (8.0mm), Nitrofurantion (10.5mm), Tetracycline (9.5mm) and

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cotrimoxazole (14mm). The antibiotics that inhibited the bacterial isolates can be used in the treatment of some foodborne diseases associated with the isolates. However, development of resistance to these antibiotics could pose a problem to antimicrobial therapy.

CONCLUSION

The kitchen sponges analyzed in this work were contaminated and could cause health challenges to the uses. The risk would be lowered when sponges are kept dry, partly because bacterial growth and survival would be reduced. The use of various disinfectants in domestic washing of cooking utensils would aid in eliminating these pathogens.

RECOMMENDATIONS

Since results have shown that microbial inactivation in kitchen sponges depends on a number of factors and is largely changeable, it is therefore recommended that self-disinfecting sponges should be considered for use, reusable sponges should be dried after use and detergent washing should be employed. The use of disinfectants should be encouraged too.

REFERENCES

1. Ikawa JY and Rossen JS. Reducing bacteria in household sponges. Journal of Environmental Health. 2003;7 (14): 18-22.
2. Gupta A and Chaidez C. Cross-contamination in the kitchen; effect of hygiene measure. Journal of Applied Microbiology. 2012;105: 615-624.
3. Kusumanigrum HD, Van Putten MM, Rombouts FM and Beumer RR. Effects of dishwashing liquids on foodborne pathogens and competitive microorganisms in kitchen sponges. Journal of Food Protection. 2002;6: 17-22.

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4. Mead PS, Slutsker L, Dietz V, McCaig, LF, Bresee JS and Shapiro C. Food-related illness and death in the United states. Journal of Emerging Infectious Diseases. 2008;5: 607- 625.
5. Cogan TA, Bloomfield SF and Humphrey TJ. The effectiveness of hygiene procedures for prevention of cross-contamination from chicken carcasses in the domestic kitchen. Letters in Applied Microbiology. 2002;29:354-358.
6. Duff SB, Scott EA, Mafilos MS, Todd EC, Krilou LR and Gedders AM. Cost effectiveness of the targeted disinfection program in the household kitchens to prevent foodborne illnesses in the United States, Canada and the United Kingdom. Journal of food protection. 2003;66:2103-2115.
7. Lancette GA and Tatini SR. *Staphylococcus aureus*, In: Vanderzart C. and Splittstoessser D.F. (eds.) *Compendium of methods for the microbiological examination of foods*. APHA, Washington D.C. 1992;pp533-550.
8. Gerba CP, Akrum HT, Maxwell LY, Sifuentes DR, Hoffman DR and and Koenig DW Bacterial Occurrence in Kitchen Hand Towels. Food Protection Trends. 2014;34 (5): 312-317
9. Cheesbrough M. District laboratory practice in Tropical Countries; Cambridge university press, Cambridge UK. 2000;pp143-180.
10. Ogbulie JN, Uwaezuoke JC and Ogiehor L. Introductory Practical Microbiology. *Springfield Publishers*, Owerri, Nigeria. 1997;160pp.
11. Mattick K, Durhaman K, Dominigue G, Jorgensen F and Schaffner DW. The survival of foodborne pathogens during domestic washing up and subsequent transfer onto washing

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up sponges, kitchen surfaces and food. International Journal of food Microbiology. 2003;8:213-226.

12. Beumer RR, Te Giffel MC, Spoorenberg E and Rombouts FM. Listeria species in domestic environments. Epidemiology and Infection. 1996 117:437-442.
13. Scott E and Bloomfield SF. An in-use study of the relationship between bacterial contamination of food preparation surfaces and cleaning cloths. Letters in Applied Microbiology. 1993;16:173-177.
14. Hilton A C and Austin E The kitchen dishcloth as a source of and vehicle for foodborne pathogens in a domestic setting. International Journal of Environmental Health Research. 2000;10:257-261
15. Kotula KL, Kotula AW, Rose BF and Camp M. Reductions of aqueous chlorine by organic material. Journal of food production. 1997;60:276-282.
16. Chaidez C and Gerba CP Bacteriological analysis of cellulose sponges and loofahs in domestic kitchens from a developing country. Dairy, food environmental sanitation 2002;20.834-837.
17. Lagendijk E, Asséré A, Derens E and Carpentier B. Domestic refrigeration practices with emphasis on hygiene: analysis of a survey and consumer recommendations. Journal of Food Protection. 2008;71:1898-1904.
18. Rayner J, Veeh R and Flood J. Prevalence of microbial biofilms on selected fresh produce and household surfaces. International Journal of Food Microbiology. 2004;95:29-39.
19. Erdogrul O and Erbilir E Microorganisms on sponges. Journal of food safety. 2005;6:17-22.

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20. Enriquez C, Enriquez-Gordillo R and Gerba C. Bacteriological survey of used cellulose sponges and cotton dishcloths from domestic kitchens. Dairy food and Environmental Sanitation. 2009;17:20-24.

21. Food and Drug Administration FDA releases report of the FDA food program database of foodborne illness risk factors. Food safety issues. 2004:2:1.

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Table 1: Identified Isolates

Colonial morphology	Microscopy	Gram stain	Motility	Catalase	Oxidase	Indole	Coagulase	Lactose	Mannitol	Glucose	Isolate
Smooth large, circular and creamy colonies with outlined edges.	Cocci in clusters	+	-	+	-	-	+	A	A	AG	*Staphylococcus aureus*
Smooth colonies, shiny with grey tint in nutrient agar.	Small scattered rods	-	+	+	+	-	-	-	AG	A	*Pseudomonas aeruginosa*
Circular, smooth with entire margin mucoid and translucent colonies on nutrient agar.	Short rods single and separate	-	+	+	-	+	-	AG	A^o	AG	*Escherichia coli*

+ = positive, - = negative, AG = Acid and Gas production; A = Acid production only; A^o = Slight gas and acid production.

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307 **Table 2: Morphological identification of fungal isolate**

Macroscopic characteristics	Microscopic characteristics	Probable isolate
White colonies later turns black, reverse side is brown.	Septate hyphae, unbranched of variable length double sterigmata covers the vesicle and forms a radiate head	*Aspergillus niger*

308 **Table 3: Percentage occurrence of bacteria and fungus in the kitchen sponges.**

Isolates	No. of sponges examined	No. of isolates present	% occurrence
Escherichia coli	60	60	100
Staphylococcus aureus	60	8	13.3
Pseudomonas aeruginosa	60	26	43.3
Aspergillus niger	60	9	17.3

309 **Table 4: Total viable counts from kitchen sponges collected from Umuariaga (cfu/ml)**

SAMPLE CODE	THPC	TCPC
1	7.70×10^8	6.15×10^7
2	6.37×10^8	7.65×10^7
3	8.20×10^8	3.60×10^7

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4	6.44×10^8	5.45×10^7
5	7.50×10^8	4.75×10^7
6	7.33×10^8	7.89×10^7
7	4.00×10^8	4.40×10^7
8	8.73×10^8	6.30×10^7
9	7.55×10^8	6.60×10^7
10	6.10×10^8	5.35×10^7
11	5.71×10^8	6.75×10^7
12	5.34×10^8	3.60×10^7
13	6.94×10^8	9.73×10^7
14	9.43×10^8	6.65×10^7
15	4.37×10^8	8.63×10^7
16	8.63×10^8	5.92×10^7
17	1.73×10^9	4.77×10^7
18	7.71×10^8	7.19×10^7
19	1.46×10^9	5.77×10^7
20	6.44×10^8	5.05×10^7
Range	**4.00×10^8 – 1.73×10^9**	**3.60×10^7 – 9.73×10^7**
Mean	**8.02×10^8**	**6.11×10^7**

Table 5: Total viable counts from kitchen sponges collected from Amaoba (cfu/ml)

SAMPLE CODE	THPC	TCPC
1	5.37×10^8	6.05×10^7
2	1.77×10^9	9.75×10^7
3	7.25×10^8	7.20×10^7
4	8.68×10^8	8.20×10^7
5	1.29×10^9	1.06×10^8
6	7.85×10^8	4.65×10^7
7	8.43×10^8	4.20×10^7

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8	1.28×10^9	4.35×10^7
9	9.65×10^8	8.70×10^7
10	7.25×10^8	7.66×10^7
11	7.80×10^8	9.63×10^7
12	1.86×10^9	5.65×10^7
13	5.85×10^8	4.30×10^7
14	1.03×10^9	9.80×10^7
15	3.65×10^8	5.65×10^7
16	5.10×10^8	3.59×10^7
17	9.71×10^8	6.55×10^7
18	8.40×10^8	6.75×10^7
19	5.62×10^8	4.63×10^7
20	9.60×10^8	6.10×10^7
Range	**$3.65 \times 10^8 – 1.86 \times 10^9$**	**$3.59 \times 10^7 – 1.06 \times 10^8$**
Mean	**9.13×10^8**	**6.70×10^7**

Table 6: Total viable counts from kitchen sponges collected from Amawom (cfu/ml)

SAMPLE CODE	THPC	TCPC
1	6.55×10^8	9.50×10^7
2	9.95×10^8	7.25×10^7
3	9.76×10^8	3.55×10^7
4	7.80×10^8	6.15×10^7
5	1.62×10^9	4.05×10^7
6	6.85×10^8	5.65×10^7
7	4.65×10^8	1.01×10^8
8	9.05×10^8	7.65×10^7
9	6.13×10^8	9.76×10^7
10	4.95×10^8	8.65×10^7
11	6.79×10^8	4.37×10^7
12	6.13×10^8	3.63×10^7

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13	9.10×10^8	5.60×10^7
14	6.89×10^8	8.63×10^7
15	8.25×10^8	6.15×10^7
16	7.58×10^8	6.70×10^7
17	1.93×10^8	5.35×10^7
18	9.55×10^8	4.50×10^7
19	7.05×10^8	4.80×10^7
20	6.95×10^8	3.65×10^7
Range	**4.65×10^8 1.93×10^9**	**3.55×10^7 – 1.01×10^8**
Mean	**8.47×10^8**	**6.28×10^7**

312 **Table 7: Antibiotic sensitivity pattern of bacterial isolates (mm)**

Bacterial Isolates	Amoxicillin (25µg)	Augumentin (10µg)	Gentamicin (10µg)	Nalidixic acid (30µg)	Nitrofurantoin (300µg)	Ofloxacin (30µg)	Tetracycline (30µg)	Cotrimoxazole (25µg)
Staphylococcus aureus	14.0	20.0	7.0	10.0	21.0	21.4	19.0	20.0
Escherichia coli	22.0	23.5	12.0	22.0	8.0	20.0	22.3	21.0
Pseudomonas aeruginosa	8.5	8.0	18.5	22.5	10.5	20.6	9.5	14.0

313 Key: R = Resistant, S = Sensitivity, I = Intermediate

314 **Interpretative reference range code**

Sensitive **Intermediate** **Resistant**

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Amoxicillin (25µg)	≥ 18	14-17	≤ 13
Augumentin (10µg)	≥ 15	13-14	≤ 13
Gentamicin (10µg)	≥ 15	13-14	≤ 12
Nalidixic acid (30µg)	≥ 19	14-18	≤ 13
Nitrofurantoin (300µg)	≥ 17	15-16	≤ 14
Ofloxacin (30µg)	≥ 22	14-21	≤ 13
Tetracycline (30µg)	≥ 19	15-18	≤ 14
Cotrimoxazole (25µg)	≥ 10	11-15	≤ 19

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