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STUDIES ON PREVALENCE OF *SHIGELLA* AND *SALMONELLA* SPECIES ON THE FOMITES OF SPECIALIST HOSPITAL, SOKOTO, NIGERIA

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ABSTRACT

Hospital acquired infections related to microbes are serious problems because human lives are at risk if infected. This study was aimed at determining the prevalence and susceptibility of *Shigella* and *Salmonella* species on nurses' tables and patients' beds at Sokoto Specialist Hospital, Sokoto, Nigeria. The bacterial species were isolated and identified using standard phenotypic and molecular methods. A total of 100 swab samples were collected from nurses' tables and patients' beds using cotton swabs in four wards. From the results obtained, four bacterial species belonging to two genera were recorded; *Shigella flexneri* (21), *Shigella dysenteriae* (4), *Salmonella paratyphi* (11) and *Salmonella choleraesuis* (13). Based on wards, the infectious ward had the highest number of isolates (22%), Balaraba (11%), Sardauna (10%) and women's amenities ward had the lowest (6%). Commercially formulated antibiotics profile on all the isolates, Ofloxacin was found to be susceptible to *Shigella flexneri* (15) with highest zone of inhibition, *Shigella dysenteriae* and *Salmonella choleraesuis* each with (8) and no zone of inhibition demonstrated in Cefuroxime on the isolates. Antibiotic plates prepared in the laboratory show bacterial species sensitive to three antibiotics; Ciprofloxacine, Oflaxacine and Ampicillin. All the three drugs were bactericidal against all the bacterial isolates, except Ciprofloxacin which was bactericidal against *Salmonella paratyphi*. Sokoto Specialist Hospital should improve hygiene through proper hygiene to reduce the burden of isolated nosocomial infections as *Salmonella* and *Shigella* are known to cause shigellosis and salmonellosis diseases in humans. This will protect public health by reducing hospital-acquired infections.

Keywords: Hospital, *Shigella*, *Salmonella*, Antibiotics, Prevalence, Beds, Tables.

INTRODUCTION

Hospital is buildings designed to diagnosing and treating sick and injured or preserve corpses for a period of time. However, nosocomial microbial-related infections often have increased mortality rates and are a serious problem (Bereket *et al*., 2012). These infections are spread through human interaction and the daily use of fomite materials found in hospitals, offices, schools, restaurants, games field and arena like bed shirts, tables, chairs, lockers, toilet seats and door handles and so on (Bright *et al*., 2010; Lopez *et al*., 2013). The species of *Salmonella* and *Shigella* are among the pathogens that cause hospital acquired infections known as nosocomial infections (Lincy *et al*., 2016). These infections are acquired by individuals within 48 - 72hours of admission to the hospital. However, visitors or relatives taking care of their sick people that stayed for three to five days are in the risk of being infected and other types of clinical facilities may also contribute to infection (Benenson, 1995; Ducel *et al*., 2002). The hospital surroundings are potential reservoirs of bacterial pathogens as it is a place where patients with diverse pathogenic microorganisms, a large number of susceptible immune compromised individual and materials (such as mattress, beds shirts, nurse tables etc.) are found mingling from one another during the process of interactions (Ducel *et al*., 2002). The increased frequency of

bacterial pathogen in hospital environment could be associated with rise in types of nosocomial infections.

Bacterial pathogens that can able to survive in the hospital environment for long period of time and resist disinfection are particularly more important for nosocomial infections. Bacterial pathogens isolated from hospital environments are also known to develop resistance to some antimicrobial agents (Lincy *et al*., 2016). The hospital environment can play an important role in the transmission of many diseases. It is therefore important to identify these environmental surfaces that are rich in bacteria and can harbor pathogens in hospital environments (Aschalew and Gelaw, 2011).

The global emergence of drug-resistant Salmonella and *Shigella* spp as well as many other ß-lactamase species has become a major treatment problem worldwide (Teshome *et al*., 2019). Multidrug resistant strains of Salmonella and *Shigella* spp are increasingly reported, especially in developing countries (Teshome *et al*., 2019). Antibiotics are the mainstay for treating bacterial infections. Since the discovery of antibiotics and their use as antimicrobial agents, the medical community believed that this would eventually lead to the eventual eradication of infectious diseases (Harbottle *et al*., 2006). However, overuse of antibiotics has become a major factor leading to the emergence and dissemination of multidrug resistant strains of several groups of microorganisms (Harbottle *et al*., 2006). Treatment of Salmonella and *Shigella* is complicated by the development of multidrug-resistant resistant strains, but prevention in high incidence areas relies on appropriate hygiene techniques and adequate sanitation facilities (Sansonetti, 2001).

Microbial contamination of hospital surroundings, especially wards, theatre rooms and fomites have continued to increase**s** the prevalence of nosocomial infections, resulting in high human morbidity and mortality (Singh *et al*., 2013). Due to the activities of visitors and relatives in the hospitals, there is need to identify the types of nosocomial pathogens and the possible infections caused, possible solutions to preventing these problems at Sokoto Specialist Hospital Sokoto so as to safeguard the health of the public, especially patients, hospital staff and visitors. This research work aimed to studies the prevalence and susceptibility pattern of *Salmonella* and *Shigella* species on fomite materials (nurses' tables and patients' beds) at /'

MATERIALS AND METHODS

Study Area

The study was conducted in lying wards of Sokoto Specialist Hospital, Sokoto State, Nigeria. The hospital is located at Sultan Abubakar Road, Sokoto South Local Government Area, in the capital city and lies between latitude of 13°4'0"N and longitude 5°13'60"E. Based on Nigeria population census of 2006, the state is ranked number 16 in population.

Preparation of Media

The media used for this study were Nutrient Agar, Nutrient Broth and Salmonella Shigella Agar. All the media used were prepared according to the manufacturer's instructions (Oxoid).

Collection of Samples from the Hospital

Cotton-tipped swabs moistened with sterile water were used to swab the surface of the fomites object (nurses' tables and patient beds) in four Wards of the Sokoto Specialist Hospital. Samples from thirty (30) tables and seventy (70) beds of Sokoto Specialist Hospital were collected using swab sticks from four wards namely; Female Amenity ward (26), Sardauna ward (24), Balaraba ward (24) and Infectious diseases ward (26). All collected samples were transported to Department of Microbiology Laboratory, Faculty of Science, Usmanu Danfodiyo University, Sokoto before 7am for further analysis.

Isolation and Identification of *Salmonella* and *Shigella* Species

Each swab sticks was transferred into 10mL nutrient broth (Oxoid, UK) and incubated at 37°C for 24hrs. A loop full of culture was transferred onto *Salmonella Shigella* Agar (SSA) plates using sterile wire loops incubated at 37°C for 24hrs. A loop full of culture medium was transferred onto nutrient agar using sterile wire loop and incubated for 24hrs at 37°C by repeated streaking to obtained pure strains of *Salmonella* and *Shigella* species. The bacteria were

identified by observing the color, growth, size and elevation of the colonies as described by Cheesbrough (2006).

Biochemical Characterization of Bacterial Isolates

The biochemical tests performed were Simmon citrate test, Indole test, Methyl Red test, Voges Proskauer test, Triple Sugar Iron Agar test and identification was done using Bergey's bacterial Identification Manual as described by Cheesbrough (2006).

Molecular Characterization of *Salmonella* and *Shigella* species

DNA Extraction

The phenotype-confirmed *Salmonella* and *Shigella* species were sub-cultured on nutrient broth in 15mls eppendorf tubes and incubated for 24 hours at 37^0C. The 24 hours suspension was centrifuged at 5000 rpm for 10 minutes. The sediments were transferred to a 1.5ml eppendorf tubes containing 200µL of molecular grade water. The tubes were placed in a water bath at 100^0C for 30 minutes and were centrifuged at 13000rpm for 5 minutes. The supernatant was carefully transferred into a newly labeled eppendorf tubes and preserved at -20^0C until used. The extracted DNA was quantified using a spectrophotometer and the absorbance length at 260nm and 280nm were determined using a UV-Spectrophotometer 2005 SELECTA©. The purity of the samples was determined by dividing the absorbance at 260nm and 280nm (Corkill and Raphley, 2008).

16S rRNA gene Amplification

The extracted DNA was used as a template in the amplification of the 16S rRNA. The reaction mixture for each sample was taken in 25ul containing: 12.5µL of PCR master mix (Biolabs©), 5.5µLof Nuclease free water (Biolabs©), and 5µL of DNA template and 2µLsequence of primers synthesized from Integrated DNA technology (IDT©), USA. The amplification was performed in an Applied bio system 9700 thermo-cycler according to the following procedure: Initial denaturation at 95^0C for 5 minutes, followed by a 35 cycles of: Denaturation at 94^0C for 1 minute, annealing at 55 ^{0}C for 1 minutes, extension at 72^0C 1 minute 30 seconds and final extension at 72^0C for 10 minutes (Drancourt *et al*., 2000; Mignard and Flandrois, 2006).

Agarose Gel Electrophoresis

Five (5µL) of each amplicon was mixed with 2µL of the gel loading dye (Biolabs©) and loaded on a 2% agarose gel on an electrophoresis tank containing 50ml of stained with Etydiumbromide (Sigma©). The voltage was set at 100V for 45 minutes. After 45 minutes the gel was transferred to a Biorad© gel documentation device for viewing (Sambrook *et al*., 2001).

Sequencing

Sequencing was performed according to the DNA manufacturer's instructions. The sequence was aligned with corresponding sequence of 16S rDNA and multiple alignment were generated by the cluster w program as stated by Madigan *et al*. (2000).

Antibacterial Susceptibility Testing

All *Shigella* and *Salmonella* strains were tested for sensitivity to different antibiotics using the paper disc diffusion method. A 0.5 McFarland standard bacterial suspension in 5mL of phosphate-buffered saline (Oxoid) were prepared and applied to the entire surface of Mueller-Hinton Agar (MHA) using a sterile swab stick. The inoculated plates were left at ambient temperature for 3 - 5 minutes. Using an automatic dispenser (Oxoid), a set of 8 antibiotic discs with the following concentrations were placed onto the surface of the plate containing the growth medium; Ofloaxacine 5µg, Augmentin 30µg, Erythromycin 30µg, Clotrimazole 30µg, Cefuroxime 30µg, Gentamycin 10µg, Ceftazidime 30µg and Ceftriazone 30µg. The antibiotic discs were placed individually on the surface of the agar medium using a sterilized forceps and pressed gently before incubates all the plates at 37°C for overnight. The clear zones created due to antimicrobial inhibition of bacterial growth were measured to the nearest millimeter using a ruler (NCCL, 2000).

Determination of Minimum Inhibitory Concentration (MIC)

To determine the minimum inhibitory concentration of the antibiotics to which the bacteria were susceptible, ten (10) test tubes were used. The first test tube contains 10mls of sterile nutrient broth while the remaining test tubes contain 5mls of sterile nutrient broth. Test tubes 1-9 were inoculated with microorganism, while test tube 10 contains only sterile nutrient broth. Test tubes 1 - 8 were inoculated with isolates bacterial species and antibiotics. The first test tube serves as positive control. Test tube 10 was used to check where minimum inhibitory concentration started. Test tube 9 was used to check whether the organism inoculated were still alive or not. 5mls was transferred from test tube 1 -9 and discarded five (5) mls from the test tube nine (9) making all the test tube to contain 5mls each (Cheesbrough, 2002). All the test tubes were incubated for 24hrs at 37°C to determine the microorganism tested that does not demonstrate visible growth.

Determination of Minimum Bactericidal Concentration (MBC)

To determine the minimum bactericidal concentration, all the test tubes showed no growth were inoculated into fresh nutrient agar (oxoid). MBC was determined by taking a loopful from each negative (no growth) tube in the MIC assay and inoculated onto fresh nutrient agar (oxoid). The plates were incubated at 37±1ºC for 24 hours after which they were observed for growth or otherwise of the test organisms (Cheesbrough, 2002). Descriptive statistics was used to express the data obtained.

RESULTS

From the results obtained, four bacterial species belonging to two genera were isolated namely *Shigella flexneri*, *Shigella dysentariae* and *Salmonella paratyphi* A from four ward of the Sokoto Specialist Hospital as shown in Table 1. Infectious diseases ward had the highest number of *Salmonella* and *Shigella* (22%), followed by Balaraba ward (11%), Sardauna ward (10%) and Female amenity ward had the lowest number (6%) as presented in the table 1. Table 2 shows the morphological and biochemical characteristics of the bacterial isolates. All were Gram negative bacteria due to the presence of bile salts that inhibit the growth of Gram positive bacteria. However, Table 3 shows the sensitivity test of commercially prepared antibiotics had the highest inhibition zone (15mm) and the lowest inhibition zone (8mm), while bacteria were resistant to the drug, other show no zones of inhibition. Furthermore, Table 4 presented the sensitivity test of the laboratory prepared antibiotics with the highest zone of 17mm in Ciprofloxacine on *Salmonella paratyphi* A, followed by *Salmonella choleraesuis* while *Shigella dysenteriae* and *Shigella flexneri* had the same zone of inhibitions (12). The Minimum inhibitory concentrations of bacteria and bactericidal isolates showing different growth or no growth at certain concentrations used as showed in Table 5 & 6.

Table 1: Frequency of occurrence of *Salmonella* and *Shigella* isolates on fomites

Bacterial Species	**Wards (%)**				**Total**
	Balaraba Ward	**Female Amenity Ward**	**Infectious diseases hospital**	**Sarduna ward**	
Shigella flexneri	5(45.4)	2(33.3)	10(45.5)	4(40)	21
Shigella dysentariae	0(0)	0(0)	3(13.6)	1(10)	4
			***Salmonella* species**		
Salmonella paratyphi A	3(27.3)	2(33.4)	3(13.6)	3(30)	11
Salmonella choleraesui	3(27.3)	2(33.4)	6(27.3)	2(20)	13
Total	11(100)%	6(100)%	22(100)%	10(100)	49

Table 2: Morphological and Biochemical Characteristics of the Bacterial Isolates

Glu G/r	Shape	IND	SUC	CIT	MOT	MR	VP	H_2S	GAS	LAC	ORGANISMS
_	Rod	_	_	+	_	+	+	_	_	+	-	*Salmonella paratyphi* A
_	Rod	_	_	+	_	+	_	+	_	+	+	*Eschericia coli*
+	Rod	_	_	_	+	+	+	_	-	+	+	*Salmonella cholerausuis*
+	Rod	_	_	_	_	_	+	_	_	+	+	*Shigella flexneri*
+	Rod	_	+	_	_	+	+	_	_	_	_	*Shigella dysenteriae*
+	Rod	_	+	+	-	+	+	-	+	+	_	*Proteus vulgaris*

Glu = glucose, G/r = Gram reaction Ind = indole, Suc = sucrose, Mot = motility, MR = Methly red, H_2S = Hydrogensulphide, Lac = lactose, Vp = Voges-proskauer

Table 3: Antibacterial Activity of Commercial Antibiotics against some Selected Bacterial Isolates from the Fomites

Antibiotics		**Diameter of zones of inhibition (mm)**			
	Grams	***Shigella Flexneri***	***Salmonella choleraesuis***	***Shigella dysenteriae***	***Salmonella paratyphi A***
Ofloxacine	5	15	8	8	11
Augmentine	30	R	R	R	R
Erythromycine	30	R	R	R	R
Clotrimazole	30	R	R	R	R
Ceftriazone	30	R	R	R	R
Cefuroxime	30	R	R	R	R
Gentamycine	10	R	R	R	R
Ceftazidime	30	R	R	R	R

R = Resistant

Table 4: Antibacterial activity of Laboratory prepared antibiotic disc on Bacterial isolates from the Fomites

Antibiotics	**Gram**	**Diameter of zones of inhibition in mm on the isolate**			
		Shigella Flexneri	***Salmonella choleraesuis***	***Salmonella paratyphi A***	***Shigella dysenteriae***
Ampiciline	10	R	I	R	R
Amoxiciline	30	R	I	S	R
Tetracycline	10	R	S	I	R
Ofloaxacine	5	I	I	I	R
Chloramphenicol	10	R	S	I	S
Ciprofloxacine	5	I	I	I	I

R = Resistant, S = Sensitive, I = Intermediate

Table 5: Minimum Inhibitory Concentration of Bacteria Isolated from the Fomites

Tested Organisms	**Ciprofloxacine/Concentration in µg/ml**						
	40	20	10	5	2.5	1.25	0.625
Salmonella choleraesuis	-	-	*	+	+	+	+
Salmonella paratyphi A	-	-	-	*	+	+	+
	Ofloxacine /Concentration in µg/ml						
	20	10	5	2.5	1.25	0.625	0.3125
Salmonella paratyphi A	-	-	*	+	+	+	+
	Amoxicilline/ Concentration in µg/ml						
	60	30	15	7.5	3.75	1.75	0.875
Salmonella choleraesuis	-	+	+	+	+	+	*

+ = there is growth, - = No growth, * = MIC

Table 6: Minimum Bactericidal Concentrations of Bacteria Isolated from the Fomites

Tested Organisms	**Ciprofloxacin/Concentration in µg/ml**			
Salmonella choleraesuis	40	20	10	5
Salmonella paratyphi A	-	-	-	*
	Ofloxacin/concentration in µg/ml			
	20	10	5	
Salmonella paratyphi A	-	-	*	
	Amoxyciline/concentration in µg/ml			
	60	30		
Salmonella choleraesuis	-	*		

+ = there is growth, - = No, * = MBC

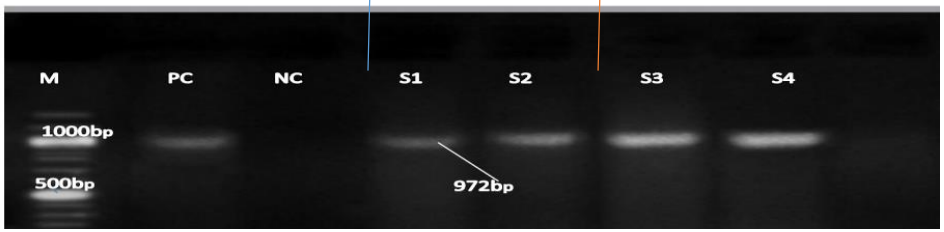

Figure 1: Agarose gel image of the 16s RNA product of two selected bacteria

GAGTCYCTACACTAKTSYTTAWGCGTTAGCTGCRGCACWRAGGGGYGGAAACCCCCTAACACTTAKCACTCATCGT
TTACGGSGKGGACTACCAGGGWATCTAATCCTGTTTGCTCCCCACSYTTTCGCSCCTCAGSGYMAGTWACAGACCA
AAAAGYCSCCTTCSCCACTGGGGTTCCTCCAAATCTCTACSCWTTTCACCGYTACACTKGGAATTCCACTTTCCTCTT
CTGCACTCAARYCCCCCAKTTTCCAATGACCCTCCACGGTKGASCCGGGGGTTTTCACATCAAACTWAAAGGACCSC
CTGCSCCCCCTTTACSCCCAAAAATTCCGGACAACSCTKGCCACCTACKTATTACCGCGGYTGCTGGCACGWAKTTA
SCCGGGGTTTTCTAWTAAGGWACCGYCAAGGTACACCCAGAAACTASTGTATTTGATCTTAAATWACTACARAKTT
TTACGATCSGAAAACCTTCTGCWTTCCCCCGGTGTTGCTCWATCAGGCTTTCKCCCWTTGKGAAGAATTCCGKACT
GYTGCCTCCCSCARGAGTGTGGGCCGYCTCTCASKGCCAGTGTGCCCAATCACCCTCTSAGGWCCGYTACCTYTCCY
CSCYTTGKAGAGCCCSYTAGCTCACWAWCTAKMTAATGCGCCGCGKGCCCATCTTATAGCGACAGASAGATGCCGT

AKGAMTYGTACWCRTGCAAGTCGAGCGAGCAGGAGWWSSWKCTTGCWCCTWTGACGYKASSGRCGGACGG
GTGACTAACACGTGGGCTACCTACCCTATAGTTTGGGATAACTCCRGGGAACCGGGGCTAATACCGAATAATC
TCTTTTACTTCATGGYGAAWKACTGAAACACGGCATCTCRCTATGGCTATASGATGGGYCCGCTGSKCATTAR
CTARWTGGYGAGGYAACGGSTCRCCRMGGMGACGATCTGWAASCGACCTGAGAGGGWGAGCGACCGCACT
GSGACTGAGACTCSGMCCRGACTCCTACGGGAGGAATCTTKMRGGAATSTTGCACAATGGGYGAAACCCTGA
TGCAKGAACGCCGCRKGAKTGCWGAASGTTTTCSTATCKTAAAACTCTGTAGTAAGGSAAGACCAAGTAGAG
GAASTCACTGGCTGTACCTTGMCAGCACCTTATTAGAAACCCACGGCTAACTACGTGCCCRAACTTACTGTGA
TATWWAGTGTGCCAARCGTTGTCCKRAATTATTGATGTGWAAAGACGCGCGCARGMKCTCCTTTCAGTCTGA
TGTGATGRYCCACGGCTCAACCGTGGAGGGTCATTGGAATTCTGKGGGACTTGAGWGMATAMSAGKAMAKT
GGAATTCCAAKTGTAGCGATAAAATGCGTAKATGATTYTGTAWCTGCACCWGTGGSTGAYGGYRACTTWTCT
GAWCTGTCAGGTGACACATSAGGCWTSGAARTCCMCGGGGGAKCAACACATSGATTAMTATACCCTGSGYKR
TSCACWTCGCGYAYAYGATGCASTGCTAWGYGTTAKRGGGKATTTYCCGYCCCSKTTRKTSCCYGSTRGMTAA
CKWCWTTASKCWCWCKGMCWTKGSGRAGTAACCKKTMCACKAGAYTGAAATYTCAAAKGARATTTTGMSC
G

Figure 3: DNA Sequence of Forward Primer of *Shigella flexneri*

GGMCYCYCYAKAAGRATCTATATGCGTTAGCTGCRGCACTAAGGGGYGGAAACCCCCTAACACTTAGCACTCAT
CGTTTACGGCGKGGACTACCAGGGTATCTAATCCTGTTTGCTCCCCACGYTTTCGCGCCTCAGCGTCAGTTACAGA
CCARAAAGYCSCCTTCSCCACTGGTGTTCCTCCAAATCTCTACGYWTTTCACCGCTACACTWGGAATTCCACTTTC
CTCTTCTGCACTCAAGWCCCCCAGTTTCCAATGACCCTCCACGGTKGAGCCGGGGGYTTTCACATCAMACTTAAA
RGACCGCCTGCSCSCSYTTTACSCCCAAWAATTCCGGACAACGYTTGCCACCTACGTATTACCGCGGYTGCTGGCA
CGWAGTTAGCCGKGGYTTTCTMMTAAGGYACCGTCAAKGTACACCCAGAAACTACTGTTTTTGWTCTTCCMTTA
CAACAGAGTTYWACGATCSGAAMACCTTCTTCWTTCCCCCGGTGGTGCTCCATCCCGCTTTCSCCCMTTGYGRAA

Figure 2: DNA Sequence of Reverse primer of *Salmonella typhimurium*

AGACKGAGTRACTTGMAGTCGAGCGACAGAAAAGGAGCTTGCTCCTTTGACGTKAGCGGCGGACGGGTGASTAACACGTG
GGCAACCTACCCTATAGTTWGGGATAACTCCGGGAAACCGGGGCTAATACCGAATAATCTCTTTTGCTTCATGGWGAAAGA
CTGAAAGACGGWTTCGGCTGWCGCTATAGGATGGGCCCGCGGMGCATTAGCTAGKTGGYGAGGTAACGGCTCMCCAAGG
CGACSATGCGTAGCCGACCTGAGAGGGYGATCGGMCWCACTGGGACTGAGACACSGYCCRGACTCCTACGGGAGGSAGCA
KTRGGGAATCTTCCACAMTGGGCGAAAGCCTGATGGAGCAACGCCGCGTGAGTGAMGAAGGTTTTCGGATCGTAWAACTC
TGYTGTAAGGGAAGAACAWGWACRGTASTAACTGGSTGTACCTTGACRGYACCTTATTAGAAAGCCACGGCTAACTACGTG
CCACCAGACGCGGYAATACGTAGGTGGCAAGCGTTGTCCGGAATTATTGGGCGTAAAGCGCGCGCATGCGGTCCTTTAAGT
CTGATGTGAAWGYCCACGGCTCARCCGTGGAGGGTCATTGGAAACTGGKGGACTTSAGTGCRGARGAGGAAAGTGGRATT
MCAAGTGTAGCRGTGAAATGCGTAGAGATTTGTAGGAACACCAGTGGCGAAGGCGACTKTTCTGRACTGYAASTGACGCTS
AGGCKYGAAASCSYGGGGAKCAMACASGATTAGATACCCTGGTAGTSCACGCCGTAAACGATGAGTGCTAAGTGTTMGGGG
GTYTCCCGCCCCKTAGTGCTGCRGMTAACRCATTAAGCACTCMGTCTGGGGGAGTACGGTCRCAAGACTGAAACTCAATGA
ATTTGACCGG

Figure 4: DNA Sequence of Forward Primer of *Salmonella typhimurium*

Noted: *Salmonella paratyphi* A, *Shigella dysenteriae* did not pass the sequencing quality control test. The result of bacteria that pass the sequencing quality control test can be seen in the figure 1. The sequence analysis of 16_S

DISCUSSION

Infections due to fomites interactions in humans vary in severity and significance, but understanding the molecular, prevalence of bacteria and monitoring antibiotic resistance is essential in curtailing hospital-acquired infections around the World. From the results obtained, four bacterial species belonging to two genera were isolated and identified namely; *Shigella flexneri*, *Shigella dysentariae* and *Salmonella paratyphi* A on nurses' tables and patient beds in four ward (Balaraba, female amenity, infectious diseases and Sadaurna ward) of Specialist Hospital Sokoto similar species were reported by Oweseni *et al*. (2021) from hospital environment. These bacterial species isolated from 30 nurses' tables, 70 bed sheets and mattresses were not surprise because, Seguija *et al*. (2016) reported the presence of microorganisms on different beds and door handlers in Kiwoko Hospital, Uganda. Additionally, Lincy *et al*. (2016) in India hospitals have isolated and identified microorganisms from door of public restrooms. Most prevalence bacterial isolates in this study were *Shigella* species with 25%, higher than the reports of many researchers around the World Ashenfi and Gedebou (1985) (9%), Asrat *et al*. (1999) (11.7%) in Tikur Anbessa, Ethio-Swedish Children's Hospital, Reda *et al*. (2011) (6.7%). On the other hand, Lincy *et al*. (2016) recorded the prevalence of *Salmoniella* and *Shigella* with (72.72) each which is higher than that in our study. The higher prevalence of Shigella genera recorded in the study could be attributed to improper public awareness on personal and environmental hygiene by various stakeholders in health and service facilities as well as human occupations. Many studies have showed that microbial diversity were different in the same surroundings such as schools, homes and hospitals (Rintata *et al*., 2008; Tringe *et al*., 2008). That is the reason why rooms in the same house contain different microorganisms due to the human activities, the ecological nature that favors the distribution/growth of microbes as well as materials inside.

The PCR products of isolated bacterial strains were sequenced using the Sanger sequencing method. The sequence data of the 16S rDNA of the bacterial isolates were subjected to phenotypic analysis. These were supported by the statement of Corkill and Raphley (2008), justified that, 16S rDNA gene sequences provide accurate grouped of the organisms, even at subspecies. This method might be considered as a tool for rapid identification of hospital acquired infection bacteria and other research areas.

The infectious diseases ward in our research had the highest number of bacterial species (22%), Balaraba ward (11%), Sardauna ward (10%) and the female amenity had less (6%). These differences in prevalence could be due to the fact that majority of the patients are literate with high economic status and have access to balance diet that makes them less susceptible to this bacterium. Furthermore, in female amenity (6%) no occupancy of patients, one person per room and regular sanitations is observed time to time per day. But, in the Infectious diseases ward all the patients met during the study were illiterates, poor and too much congested and poor sanitation of the room, which may be the cause of the high rate of bacterial species. These statements are consistent with a previous study conducted by Aziz *et al.* (1990), education is vital to create public awareness of the mechanisms for managing infectious microorganism and controlling other factors that lead to disease. Poor environmental sanitation, malnutrition, inadequate water supply, poverty, weak immune system and limited education are the major factors in the occurrence, spread and severity of hospital acquired infections. There was widespread of *Salmonella* and *Shigella* in the Specialist Hospital Sokoto. Recently, a number of human disease outbreaks involving *Salmonella* and *Shigella* have caused many deaths in recent years in developing countries (Mengistu *et al*., 2014).

Minimum inhibitory concentrations of the drug were 10µg, 5µg, 5µg against *Salmonella choleraesuis* and *Salmonella paratyphi* A. However, commercially prepared antibiotic plates showed that all the bacterial isolates were sensitive to Ofloxacin only (Table 2). This is harmony with the statement of Onoalopo *et al*. (2015) who stated that, the antibiotic susceptibility profile of the *Staphylococcus aureus* isolated from the sampled door handles showed 76.6% resistance to Ofloxacine. Also analogy with the report of Otokunefor *et al.* (2020) who revealed very high levels of resistance (80–100%) against 75% of the test antibiotics on the enterococci bacterial species. Therefore, microorganisms become resistant to a drug when they regrow within the zone of inhibition due to the selective pressure created. According to Lincy *et al*. (2016) bacterial species were resistant to at least one specific antibiotic this parallel with findings. In addition, high rates of antibiotic-resistant bacteria may be due to inappropriate or uncontrolled use of antibiotics. Furthermore, high antibiotic susceptibility of *Salmonella* to ciprofloxacin in this study was also reported by Mengistu *et al*. (2014) in Ethiopia. The resistance of *Salmonella paratyphi* A to ampicillin and tetracycline was consistent with the results of Beyene and Tasew (2014), in which most

of the *Salmonella* isolates were resistant to ampicillin. In the current study, multidrug resistance to seven drugs was observed in *Salmonella* and *Shigella* from commercially formulated antibiotics. *Shigella* isolates were susceptible to ciprofloxacin. The resistance of *Shigella* spp. towards ampicillin and tetracycline was in agreement with studies conducted by Roma *et al.* (2000) who reported high rate of resistance of *Shigella* spp. to ampicillin (93%), erythromycin (90%), tetracycline (90%), and cotrimoxazole (56%). An increasing resistance of *Salmonella* and *Shigella* bacteria makes the treatment of infections more challenging and difficult. Thus, epidemiological information and monitoring systems are necessary to control *Salmonella* and *Shigella* infections in the public health sector.

CONCLUSION

Based on the findings, it was concluded that, four bacterial species belonging to two genera were prevalence in nurses' tables and patient beds of Specialist hospital Sokoto State that are potential mean of transmitting Hospital-acquired infection diseases among patients, medical staff, visitors and workers. However, sensitivity test conducted using commercially prepared disc showed that Ofloaxacine was sensitive *Salmonella a*nd *Shigella* species. Laboratory prepared antibiotic plates; shown bacterial species were sensitive to three antibiotics (Ciprofloxacine, Oflaxacine and Ampicillin).

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