ISOLATION AND DETERMINATION OF ANTIMICROBIAL RESISTANCE PATTERN OF PATHOGENIC BACTERIA IN SINKS WITHIN SURGICAL THEATERS OF KORLE-BU TEACHING HOSPITAL.

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ABSTRACT

A sink is a basin-like structure with plumbing for water supply and drainage. It can be made from variety of materials including stone, stainless steel, copper, ceramic etc. to suit the environmental conditions and purpose of use. In the hospital, sinks are usually used for washing and drainage. Hospital sinks sometimes serve as reservoirs of pathogens that cause several nosocomial illnesses amongst admitted patients. The aim of the study was to evaluate the importance of hospital sinks as reservoirs of nosocomial pathogens. A total of 28 surgical theater sinks in Korle-Bu teaching Hospital (KBTH) were swabbed. Primary cultures were performed on blood agar plates for each swab. McConkey agar plates were used to subculture growth colonies from primary cultures. The agar plates were incubated at 37^0C for 24 hours overnight. Standard biochemical tests as well as gram staining were then performed to identify the bacteria isolates. Bacteria resistance was determined using the Kirby-Bauer disc diffusion technique. The data obtained was statistically analyzed using Statistical Package For Social Sciences (SPSS). The prevalence of organisms in sluice and hand-scrub sinks from 12 surgical theater sinks at the 5 study sites were 41 (44.6%) and 51 (55.4%) respectively. Bacterial load obtained from the study sites were 58 (63.0%), 15 (16.3%), 8 (8.70%), 8 (8.70%), and 3 (3.30%) for sinks at the Surgical Block, Accident Center, Maternity Block, Gynaecology Department, and National Cardiothoracic Center respectively. There were 92 total bacterial growth; 43 (46.7%) were *Pseudomonas aeruginosa*, 16 (17.4%) were *Bacillus spp.,* 13 (14.1%) were *Kiebsiella pneumoniae*, 18 (19.6%) were *Enterobacter spp.*, and 2 (2.2%) were *Staphylococcus aureus*.

Multiple drug resistance was observed among the various bacteria isolated. Pathogenic bacteria growths were observed in sluice and hand-scrub sinks within surgical theaters of the Accident Center, Gynaecology Department, Maternity Block, National Cardiothoracic Center and the Surgical Block in KBTH.

Key-Words: Sinks, Nosocomial, Resistance, Pathogenic, Bacteria

INTRODUCTION

The many benefits of hospital sinks includes hand sanitization and hence the reduction of nosocomial infections (AHRQ, 2001). Sinks are also used for washing and rinsing surgical wares such as forceps, clamps, scalpels, suction tips etc. Sometimes, designated sinks can be used to drain body fluids in the hospital. In addition, sinks have maximum containment enough to prevent splashing to equipment and patient area (Fletcher and Fletcher, 1902).

There are however, some disadvantages associated with the use of sinks in hospital settings especially in surgical theaters. These challenges raise major health concerns. Conditions that permit the growth of pathogenic microbes especially *Pseudomonas aeruginosa* sometimes exist in hospital sinks (Ayliff *et al.,* 1974). Infested sinks were the sources of *Klebsiella oxytoca* outbreak in the series of outbreaks that infected about 66 patients between 2006 and 2011 in Toronto's Mount Sinai hospital (Press, 2012). A hospital sink was also the source of fatal *Legionella pneumophila* infection in a leukemia patient (Alexandre *et al*., 2008). Improperly designed and installed sinks can act as environmental receptacle for pathogenic microbes, which may lead to the outbreak of gram-negative bacilli (PIDAC, 2010).

Sink waste-traps have been proven as viable reservoirs of pathogenic bacteria especially *Pseudomonas aeruginosa;* as much as 7-5 million living bacteria were found on sink waste-traps

as well as the sides of the sink (Newsom and Cantab, 1968). When sink waste-traps are however sterilized, microbial retention is diminished (Noone *et al*., 1983). Hot water faucets on some sinks can aerosolize bacteria by producing hot running water, which heats up the washbasin, and cause steam production. The steam carries pathogens within the sink into the air. Pathogens such as *Legionella pneumophila* and *Pseudomonas aeruginosa* can be easily aerosolized (Gary *et al*., 1985). Air driers can also aerosolize water borne pathogens especially *Legionella sp* (WHO, 2009).

It is therefore imperative to carry out necessary maintenance as well as other corrective measures to ensure that pathogenic microbes are eradicated from hospital sinks and accessories in order to reduce the risk of nosocomial infection in surgical theaters as well as other wards. Sinks should be designated for hand washing alone (PIDAC, 2010). Hospital sinks must be disinfected, drained to get rid of any stagnant water or residues (normally filtered by sink waste-traps) which may harbor microbes, and sterilized regularly. Hand-wash sinks should be fitted with hot and cold-water mixer tap (with regulated temperature) that dispenses from a single outlet (SARI, 2001). Sinks must be designed with deep basins to minimize splashing, built with impervious materials such as porcelain, stainless steel etc., and must be well fitted into cabinetry and wall to prevent leakage (ASHE, APIC, 2001).

Hospital sinks serve as reservoirs of agents that cause nosocomial infections. Such agents isolated from hospital sinks include *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Burkholderia cepacia*, *Legionella pneumophila*, *proteus spp* etc. (Perryman and Flournoy, 2003). The possible health burden of nosocomial infection is heightened by the increasing trend of

antibiotic resistance in nosocomial pathogens. A study in the United States showed multiple drug resistance (MDR) crude prevalence rate of 14% for *Pseudomonas aeruginosa* (Tam *et al*., 2010). Another study in India showed MDR prevalence of 76% for *Klebsiella pneumoniae* (Sikarwar *et al*., 2011). *Klebsiella pneumoniae* isolated from the intensive care unit and long term facility of hospital units were found resistant to commonly used ß-lactams, aminoglycosides, and quinolones (DOH, 2013). Nosocomial infecctions constitute an important cause of morbidity and mortality in Ghana. It is thus necessary to know the possible role sinks may play in such infections.

Sinks are used in hospitals for washing and drainage. In surgical theaters, sinks are used by surgeons to wash their hands prior to putting on surgical gloves before surgery (Paterson *et al*., 1995).Numerous studies on reservoirs of bacterial pathogen within hospital sinks have been conducted off the coast of Ghana. *Klebsiella oxytoca* was recently isolated from a hospital sink in Toronto, Canada (Press, 2012). Gentamicin and Amikacin (aminoglycosides) resistant bacteria were found in sink drains of health related facilities (Perryman *et al*., 2003). Furthermore, *Pseudomonas aeruginosa* was detected in aerosols produced from hot water faucets in hospital sinks (Döring *et al*., 1991).

In Ghana however, no research work has been published on reservoirs of pathogenic bacteria within sinks of surgical theaters in hospitals. It is expected that such a study would promote the necessary information to help educate the public on the health hazards posed by bacteria growth in hospital sinks. Additionally, the study would provide information on the types

of bacterial pathogens haboured by sinks in the hospital and the possible role of sinks in nosocomial infection.

METHODS

STUDY DESIGN AND SITE

The study was cross sectional. Samples were collected from surgical theaters at Surgical Block, National Cardiothoracic Center (NCC), Department of Gynaecology, Maternity Block, and the Accident Center (AC) in Korle-bu Teaching Hospital (KBTH) in Accra, Ghana. The samples were analysed at the The University of Ghana School of Allied Health Sciences (SAHS) Microbiology Laboratory and the University of Ghana Medical School (UGMS) Microbiology Laboratory.

ETHICS

Ethical clearance was sought from the Ethics and Protocol Review Committee of The University of Ghana School of Allied Health Sciences. Permissions was sought from the Head of Surgical Department of KBTH, Head of Obstetrics and Gynaecology Department, Head of The Cardiothoracic Surgery Unit, and also from the Management of KBTH before the study was commenced.

INCLUSION AND EXCLUSION CRITERIA

Inclusion criteria

- Hand-scrub and sluice sinks in surgical theaters within KBTH

Exclusion criteria

- Abandoned sinks, broken sinks, and Infested sinks within surgical theaters of KBTH.

PROCEDURE FOR QUESTIONNAIRE ADMINISTRATION

A well-defined questionnaire probing for the functional, structural, and hygienic state of sinks was administered to the theater staffs to determine the suitability of the surgical sinks for the study. A cover sheet containing the title and pertinent details of the project like the aim and importance of the project as well as expectations from participants was clipped to the questionnaire.

SAMPLE COLLECTION AND PROCESSING

Sluice and hand-scrub sinks in the surgical theaters were conveniently sampled three times; the first time in the morning, second time in the afternoon, and third time in the evening. Sink waste-traps, sinkholes, and basins were sampled with sterile cotton swabs moistened with 0.85% physiological saline. The swab samples were then placed in sterile containers and transported to the SAHS microbiology laboratory for processing. Once at the laboratory, the samples were quickly cultured on Blood Agar plates and immediately incubated aerobically at 37^0C for 24 hours to grow for the possible organisms present to grow. Isolated colonies were then inoculated on McConkey agar plates and incubated overnight at 37^0C. The plates were re-incubated for 48 hours at 37^0C when no visible organism growth was observed after 18 hours of initial incubation. Any culture that showed mixed growth was sub-cultured unto Blood and McConkey agar plates for further isolation and identification.

The colony isolates from the McConkey agar plates were stained by Gram stain in order to distinguish gram-positive organisms from gram-negative organisms. Biochemical tests were performed on the isolates to identify them. Catalase and coagulase tests were performed on all gram positive cocci. Indole, citrate utilization, urease, triple sugar iron, oxidase, and motility tests were performed on all gram negative rods. 3.3.2 Antimicrobial Resistance

The antimicrobial resistance of isolated organism was determined by the Kirby-Bauer disc diffusion method (Bauer *et al.,* 1966)

DATA MANAGEMENT PLAN

Data obtained from the study site was handled with utmost confidentiality.

DATA ANALYSIS

The data derived from the laboratory investigations was analyzed statistically using the Statistical Package for Social Sciences (SPSS) version 16.0 windows software. A summary of data was presented using descriptive statistics of mean, frequency, and graphs. An analysis was done to compare the results obtained from the various units involved to assess the range of sink contamination.

RESULTS

A total of 28 sinks, 15 hand scrubs and 13 sluices, were sampled from 12 surgical theaters in 5 units namely; Surgical Block, National Cardiothoracic Center, Accident Center, Maternity Block, and Gynaecology Department. The distribution of sinks in surgical theaters were; Pediatric (3), General (3), Maxillofacial (2), Urology (2), ENT (2), Eye (2), Neurosurgery (3), CT (2), Accident (2), Orthopaedic (2), Obstetrics (3), and Gynaecology (2). The distribution

of swab specimen in the five hospital units including the culture is shown in Table 1. Out of a total of 72 Swabs of sink holes, sink drains, and sink basins analysed, 11 swabs had insignificant or no growth. The remaining 61 swabs produced significant bacterial growth and five (5) different bacteria species were identified: *Pseudomonas aeruginosa*, *Bacillus sp*, *Klebsiella pneumoniae*, *Enterobacter sp*, and *Staphylococcus aureus*.

From 92 total bacteria isolates obtained from sinks at the five (5) units, 58 (63.0%) was recorded at Surgical Block, 15 (16.3%) at the Accident Center, 8 (8.70%) at Maternity Block, 8 (8.70%) at the Gynaecology Department, and 3 (3.30%) at the National Cardiothoracic Center.

51 (55.4%) of the total bacterial growth were observed in cultures of samples obtained from the hand-scrub sinks whiles 41 (44.6%) were observed in cultured samples from sluice sinks.43 (46.7%) of the 92 bacteria growths were *Pseudomonas aeruginosa*, 18 (19.6%) were *Enterobacter spp.*, 16 (17.4%) were *Bacillus spp.,* 13 (14.1%) were *Kiebsiella pneumoniae*, and 2 (2.2%) were *Staphylococcus aureus* (Refer to Appendix D, Tables 4-8). The prevalence of bacteria growth in sluice and hand-scrub sinks in surgical theaters within the study sites has been presented in the tables below. The antimicrobial resistance pattern of the bacterial isolates has also been illustrated in the figures below.

Table 1: Swab specimens collected from sinks in various units of KBTH

Hospital Unit	**Swab No.**	**Culture Pos.**	**Culture Neg.**	**Total *n* (%)**
	***n* (%)**	***n* (%)**	***n* (%)**	
Accident	12(16.2%)	12(100%)	0(0.00%)	**12(100%)**
Cardiothoracic	6(8.11%)	4(66.7%)	2(33.3%)	**6(100%)**

Gynaecology	6(8.11%)	6(100%)	0(0.00%)	**6(100%)**
Maternity	6(8.11%)	6(100%)	0(0.00%)	**6(100%)**
Surgical	44(59.4%)	35(79.5%)	9(20.5%)	**44(100%)**
Total n (%)				**74(100%)**

Table 2: Prevalence of bacteria isolated from sinks in all surgical theaters at KBTH

ORGANISMS	**SINKS**	
	SLUICE	**HAND-SCRUB**
Pseudomonas areuginosa	18(43.9%)	25(49.0%)
Bacillus spp	10(24.4%)	6(11.8%)
Klebsiella pneumoniae	9(22.0%)	4(7.84%)
Enterobacterspp	3(7.32%)	15(29.4%)
Staphylococcus aureus	1(2.44%)	1(1.96%)
TOTAL	**41(100%)**	**51(100%)**

Key: Pos. = Positive, Neg. = Negative, No. = Number, *n* = Count

Figure 1: Percentage of *Staphylococcus aureus* resistance to some antibiotics.

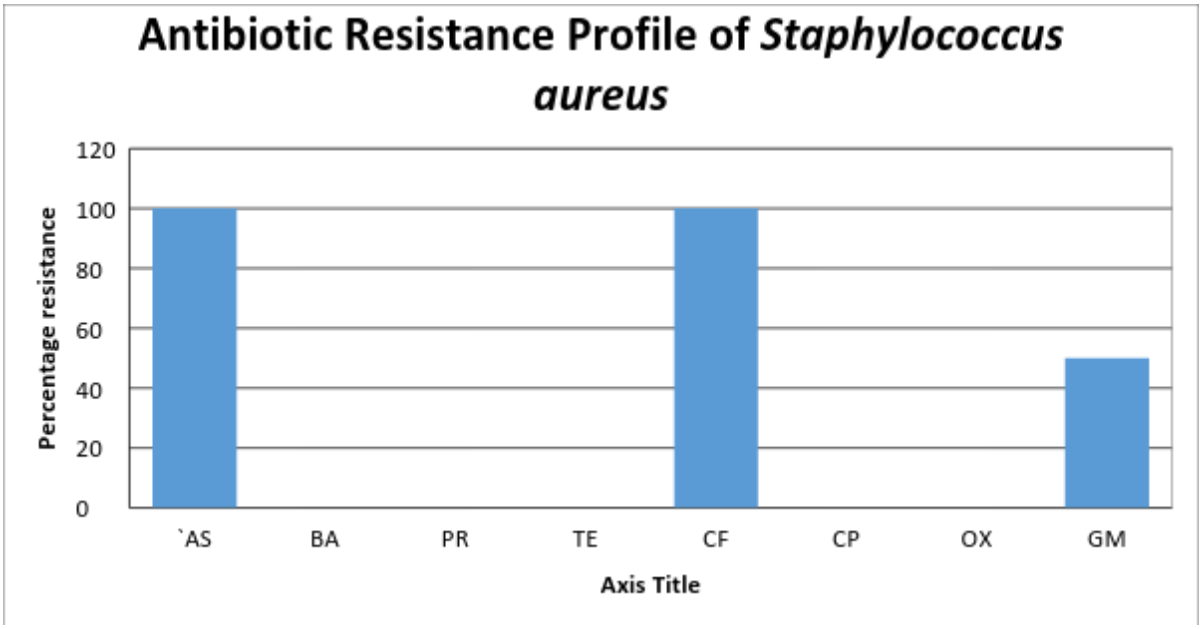


Figure 2: Percentage of *Pseudomonas aeruginosa* resistance to some antibiotics.

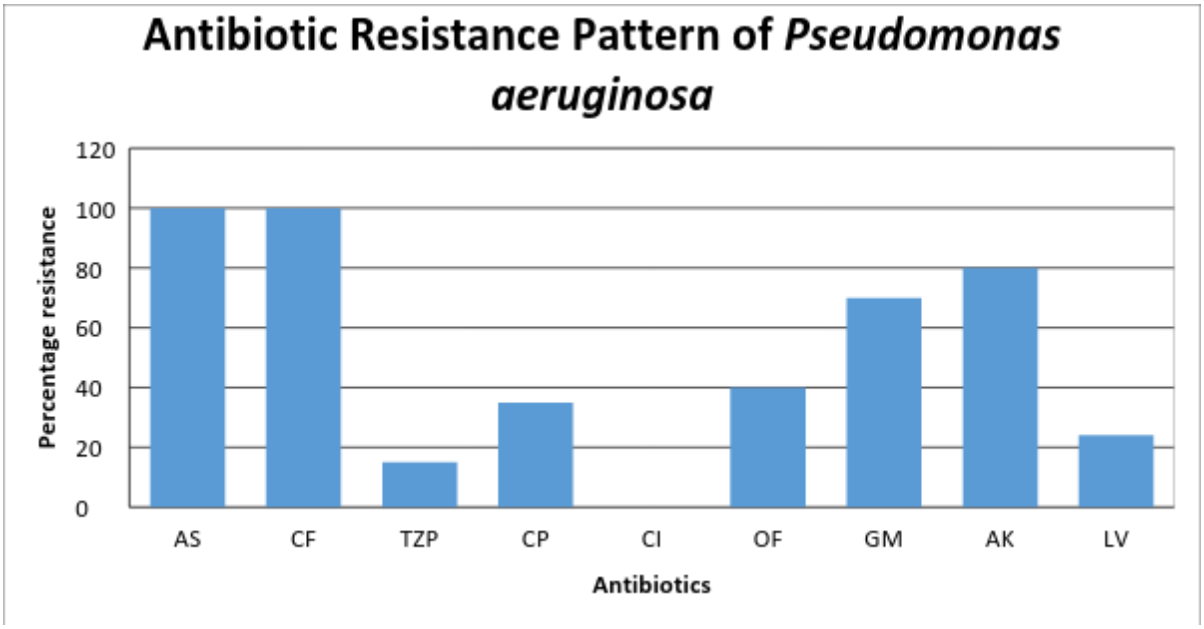


Figure 3: Percentage of *Bacillus spp.* resistance to some antibiotics.

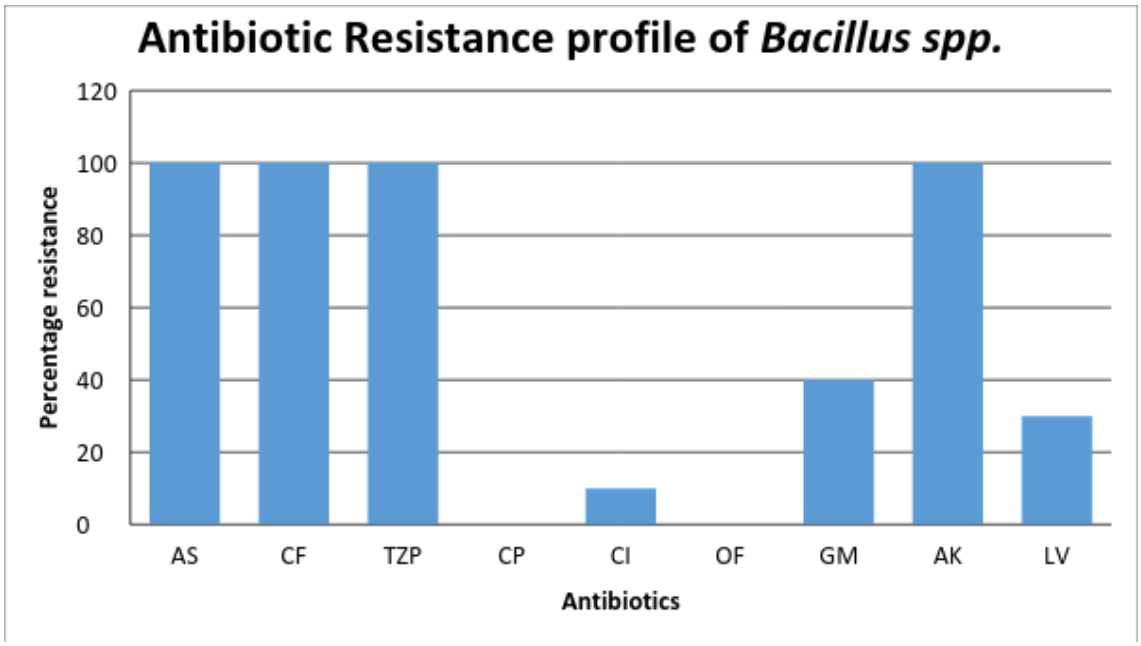


Figure 4: Percentage of *Klebsiella pneumoniae* resistance to some antibiotics.

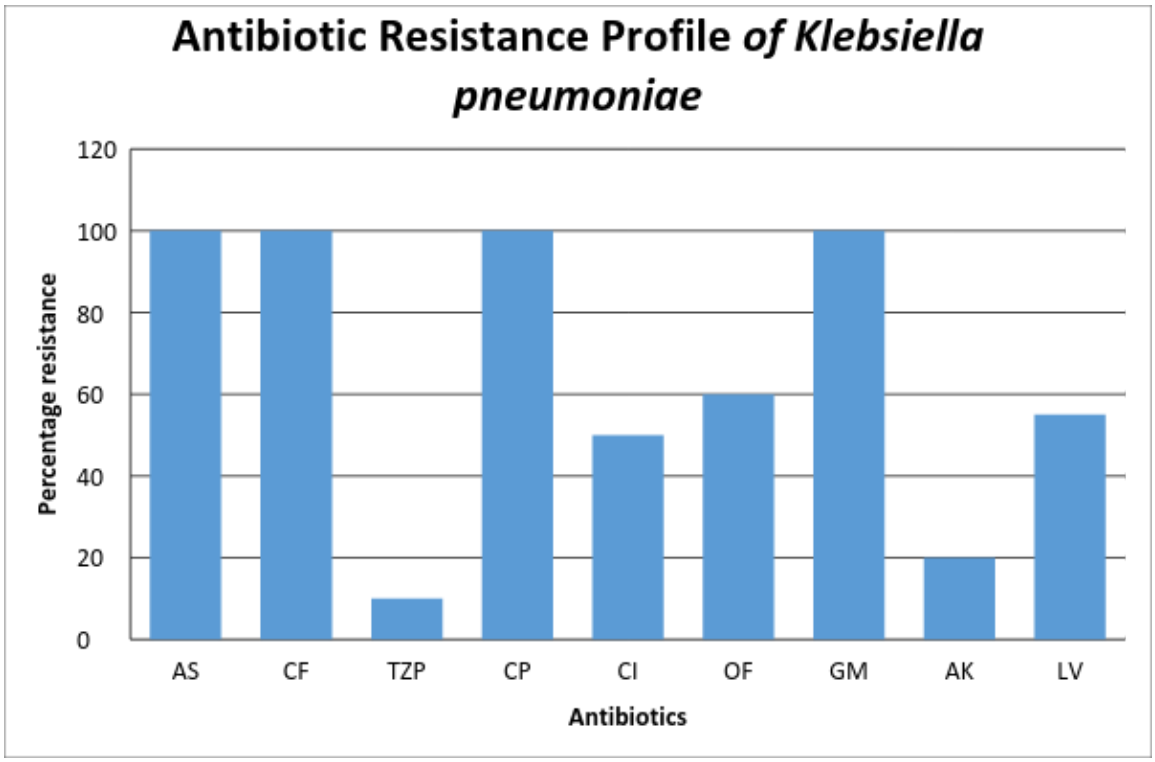
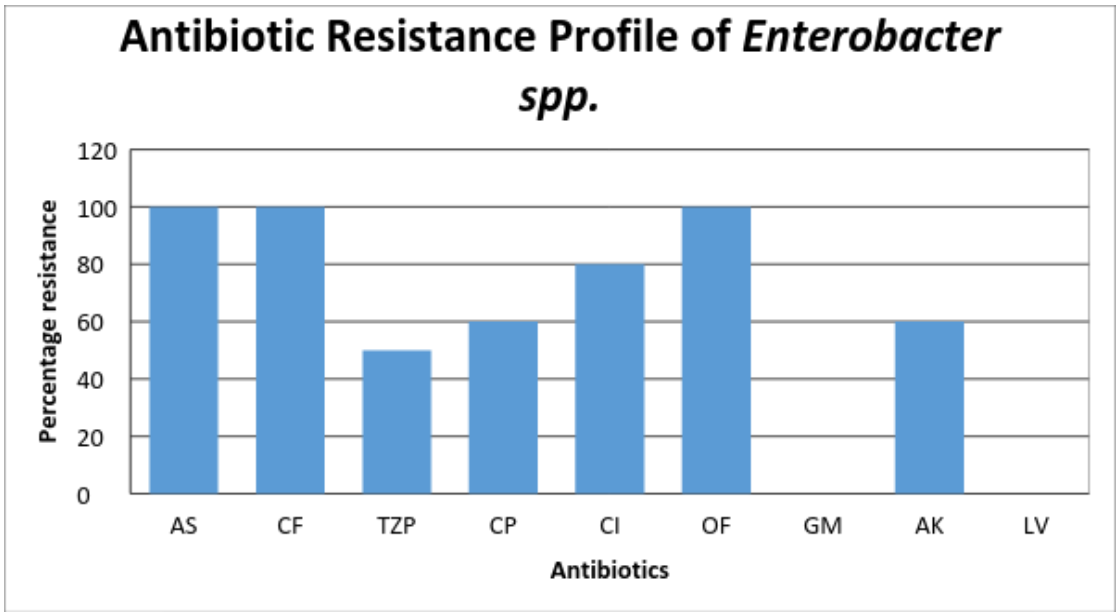


Figure 5: Percentage of *Enterobacter spp.* resistance to some antibiotics.



Key: AS =Ampicillin, AK = Amikacin, BA = Co-Trimoxazole, PR = Cephalexin, TE = Tetracycline, TZP = Tazobactam, CF = Cefotaxime, CP = Ciprofloxacin, CI = Cefuroxime, PF = Prulifloxacin, OX = Oxfloxacin, OF = Ofloxacillin, CX = Cloxacillin, RF = Roxithromycin, LM = Lincomycin, GM = Gentamycin, LV = Levofloxacin.

DISCUSSION AND CONCLUSION

DISCUSSION

51 sluice sinks and 41 hand-scrub sinks were sampled from 10 theaters at the Accident Center, Gynaecology Department, Maternity Block, National Cardiothoracic Center, and Surgical Block. The highest bacterial load was observed in sinks from surgical theaters at the

surgical block. The questionnaire administered 2nd June, 2013 revealed the sinks were not adequately cleaned and that sink drains were never changed or cleaned since they were installed. The questionnaire also revealed the exterior body and surroundings of the sinks, especially theater sinks at the surgical block, were seldom cleaned. The sinks were only changed during renovations or when broken; in combination with the poor sanitary conditions, this might have been the cause of increased bacterial contamination.

It was quite clear that in the duration of this study, most of the sinks were poorly maintained due to failure on the part of the staff to regularly clean, disinfect, and change the sink drains. This observation was however exceptionally different at the Cardiothoracic theater were the sinks were cleaned and disinfected regularly. Cardiothoracic theater sinks had the lowest bacterial load.

The Surgical Block had the highest bacteria load with prevalence of 63.0% followed by the Accident Center with prevalence of 16.3%. The Maternity Block and the Gynaecology Department both had bacteria load of 8.70% prevalence. The National Cardiothoracic Center had 3.30%, the lowest prevalence recorded in the study. The high prevalence of bacterial growth observed in theater sinks at the Surgical and Maternity Blocks may be due to the fact that health workers and trainees increasingly use the facilities during operations; this thus increases the chances of bacterial contaminants in the sinks during hand-washing, drainage of fluids, and washing of surgical tools. High bacterial prevalence values at the Surgical Block may also be due to the numerous sinks sampled within its theaters as compared to the other units with fewer sinks

installed. This explanation also holds for the NCC and Gynaecology theaters due to the relatively fewer number of sinks (as compared to Surgical Block Theater) installed.

From table 2, the sluice sinks in surgical theaters of KBTH had 41 out of 92 total bacterial growths with prevalence of 44.6%. The hand-scrub sinks had 51 bacterial growth and bacterial prevalence of 55.4%. The high prevalence of bacteria in hand-scrub sinks could be due to irregular sanitizing of the sinks, using of expired or weak cleaning solutions, and increased sink usage due to increased number of operations and trainees. The bacterial prevalence in sluice sinks, albeit lower than the prevalence in hand-scrub sinks, was expected. This is due to the drainage of fluid waste and washing of surgical tools in sluice sinks. It may also be due to the poor cleaning routine of the sink.

The prevalence of *Staphylococcus aureus* was 2.17% in sinks from all units. The organism was most prevalent at the General Surgery Theater with 3 (9.09%) growths. The bacteria belong to the micrococacceae family and causes sepsis and abscess formation (Lowry, 1998). *Staphylococcus aureus* isolated in the study was susceptible to cloxacillin. Cloxacillin resistant *Staphylococcus aureus* causes serious infections.

Bacillus was not speciated in the study but given the huge prevalence of the organism in hospitals, it may be important in subsequent studies to identify the species of the organism within the hospital environment.

Pseudomonas aeruginosa had a prevalence of 46.7% in sinks from all units. The organism was most prevalent at the pediatric theater sinks with 75.0% followed by Obstetrics

Theater with 62.5%. A Nosocomial outbreak of this pathogen in pediatric units usually involves newborns and immunocompromised patients. It can cause significant morbidity and mortality (Buttery *et al*., 1998). The high prevalence of the organism in the obstetrics theater may contribute to post-operative wound infections. Outbreak of *Pseudomonas aeruginosa* in post-operative wound infection is becoming more threatful in developing countries due to degenerating hygienic measures, mass production of ineffective antiseptic and antibiotics for treatment, etc. (Masaadeh and Jaran, 2009)

Kiebsiella pneumoniae had a prevalence of 14.1% in sinks from all units. The bacteria were most prevalent at the Gynaecology and Obstetrics Theaters with 25% prevalence in both. The organism causes nosocomial infections such as sepsis, pneumonia, meningitis, surgical site or wound infection etc. immnocompromised patients and neonates in these units are at greater risk of being infected by the bacteria. *Kiebsiella pneumoniae* can infect and cause peripartum transmission to neonates in less than 48 hours (Gaillot, *et al*., 1998)

Enterobacter spp. had a prevalence of 19.6% in sinks from all units. It was most prevalent at the Accident Theater with 57.1%. The bacteria specie is the fourth commonest cause of gram-negative bloodstream infection (Al-hasan, *et al.,* 2011). This nosocomial bacterial also causes urinary and respiratory tract infections in immune compromised patients. Patients at the accident theater are thus at risk of exposure to *Enterobacter spp.* infection.

Multiple drug resistance was observed in all bacteria isolated from the various units. Antimicrobial resistance rate of the isolated organisms were generally higher for *Kiebsiella pneumoniae*, and lower for *Bacillus spp* in Gram negative isolates. Gram-negatives were

generally resistant to Ampicillin and Ciprofloxacin. This was of less medical importance since these antibiotics are considered ineffective against *Pseudomonas,* and most enterics in modern pharmacology. Furthermore, high Ciprofloxacin resistance among 480 clinical isolates was recorded for *Pseudomonas* and other *enterobacteriacae* in a recent study (El Alstal 2005; Mosby 2006; Katzung& Bertram 2007). *Staphylococcus aureus*, the only gram-positive isolate, was generally resistant to Ampicillin, Cefotaxime, and Gentamicin. This was of less clinical significance since Ampicillin, Ciprofloxacin, and Gentamicin are considered weak antibiotics against *Staphylococcus aureus*. In a recent study, *Staphylococcus aureus* resistance to Gentamicin was shown to have occurred due to gene mutations that produced aminoglycoside 6′-N-acetyltransferase and gentamicin phosphotransferase enzymes (Dowding, 1977).

LIMITATIONS

Some sinks were inaccessible as they were located in a restricted area of the surgical theaters. This was so for at least two of the sluice sinks at the anaesthesiology rooms within theaters of the surgical block. The sample size of the study was thus affected.

RECOMMENDATIONS

The following recommendations were made to the management of KBTH, and Heads of Departments of all participating units: Hospital sinks should be frequently sanitized at least 3 times a day, morning afternoon, and evening to reduce bacterial contaminants and prevent sink related nosocomial infections. The sinks should be serviced regularly (at least once every six months) to ensure the integrity of the plumbing structures are intact. Sink plugs and drains should be replaced as often as possible. Adequate sinks should be installed to cater for increased work load and the presence of trainees. Only authentic and active disinfectants should be used in the sanitization of hospital sinks. Proper sanitary measures should be implemented and hospital staffs motivated and encouraged to follow them.

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5.2 CONCLUSION

Sluice and hand-scrub sinks within theaters at KBTH harbour various bacterial agents, most of which are nosocomial pathogens. The bacterial agents isolated from the sinks were resistant to several antibiotics especially *Klebsiella pneumoniae* which was resistant to nine (9) different antibiotics.

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