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Bacterial contamination and antimicrobial susceptibility pattern of isolates from stethoscopes at a referral hospital in Tanzania

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Abstract

Background

Nosocomial infections pose a challenge to the medical field and are a global safety issue for both patients and health care providers. In developing countries, the magnitude of the problem remains underestimated and literature is very scarce. Some nosocomial infections spread via contaminated medical equipment such as stethoscopes. No data reported from Tanzania about the potential role of stethoscopes in spreading these infections.

Methods

This cross-sectional study was conducted to determine bacterial contamination and antimicrobial susceptibility of isolates at Kilimanjaro Christian Medical Centre (KCMC) from February to April 2014. One hundred medical doctors and medical students were randomly selected to participate in this study. Structured questionnaires were used to collect demographic data and behavioural information related to the cleaning and storage of stethoscopes. Participants' stethoscopes were sampled with sterile moistened cotton swabs. Laboratory analysis was done following standard microbiological techniques in the Microbiology Unit. Data analysis was done using SPSS window version 16, and p-values of <0.05 were considered statistically significant.

Results

A total of forty-six of the stethoscopes were found to be contaminated. A total of 134 bacterial strains were isolated. Of 134 isolates, 70 were potentially pathogenic, including *S. aureus*, Klebsiella ssp, Proteus ssp, *E. coli* and *P. aeruginosa*. Eighteen percent of the *S. aureus* were methicillin-resistant.

Conclusion

We found a significant amount of bacterial contamination of stethoscopes at our hospital, a significant percentage of which is Methicillin-Resistant *Staphylococcus aureus* (MRSA). Similar to other settings, in our setting in northern Tanzania, stethoscopes are important potential vehicles for nosocomial infections.

***Key words*:** Nosocomial infections, stethoscopes, MRSA, Tanzania.

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Background

Nosocomial infections (NIs) are an important cause of infections in healthcare settings. NIs can be spread by improper hand washing, and can be carried on fomites or medical equipment [9-10]. The medical equipment used most by medical doctors and students are stethoscopes which can be contaminated by fungi, bacteria, and viruses from the hospital environments.[9] Most of the existing evidence about NIs and efforts to curb their prevalence is from developed countries where there are regular surveillance systems that contribute to national reports on NIs [11]. It is a different situation in developing countries like Tanzania where there are health care system deficiencies aggravated by poverty and high burdens of infectious diseases. Furthermore, hospital understaffing, staff overworking, hospital overcrowding and inadequate infection prevention control (IPC) protocols all likely contribute to the spread of infections within healthcare settings [12].

Due to the nature of its use, it is highly susceptible to microbial contamination. Specifically, the diaphragm contacts the patient's body and may thus be contaminated by bacteria [13]. One stethoscope comes into direct contact with numerous patients per day and disinfection after each use is not a well-established practice [14]. Hospitalized patients may become colonized by bacteria, including drug-resistant bacteria, from the hospital environment. As this population's health is already compromised by medical comorbidities, individuals are at increased risk for infections with the bacteria with which they come into contact. A systematic review on NIs in Africa by Sepideh *et al* showed the prevalence of surgical wound infections post-discharge to be 21% in Tanzania [15]. In another study from Tanzania, 19.4% of patients developed surgical site infections, 36.4% of which were picked up during post-discharge follow up [16-17]. Though the prevalence of surgical wound infections does not provide a comprehensive picture of NIs in Tanzania, it does suggest that rates may be high and also highlights the scarcity of data specifically about NIs. Evidence of surgical site infections seen in Tanzania, data is largely missing from Africa and completely missing from Tanzania on the potential role of stethoscope in spreading infection.

Therefore, our study was conducted in a referral and teaching hospital in Northern Tanzania, to identify the antimicrobial susceptibility patterns of the isolated bacteria, as well as to identify the behaviours of health care workers that are risk factors for stethoscope contamination with pathogenic organisms.

Methods

Study area and settings

The study was conducted at KCMC Referral and Teaching Hospital in Moshi, Tanzania, from February to April 2014. KCMC is a 600-bed facility that serves a catchment area of over 15 million people in northern Tanzania. The hospital has more than 450 medical students, 200 doctors and 400 nurses. Ethical clearance was secured from the Kilimanjaro Christian Medical University College (KCMUCo) Ethical Committee. Permission was also obtained from the Executive Director of KCMC.

Sample size and sampling technique

Convenience sampling was used to select the doctors and students enrolled in the study. A total of 100 healthcare workers were selected, of which 41 were medical doctors and 59 were medical students. After obtaining consent from each participant, a structured questionnaire was used to collect data regarding the behaviour of that participant, specifically with regard to stethoscope care. An identification number was assigned to each department; there were 62 participants from non-surgical departments and 38 participants from surgical departments. Anonymity was maintained for all participants.

Specimen collection and identification of pathogen

Specimens were collected from the surface of each stethoscope diaphragm using a sterile cotton swab moistened with 0.9% normal saline. Using sterile technique, each swab was used to inoculate one blood agar, one MacConkey agar and one chocolate agar plate. The chocolate agar plates were incubated in a 10% carbon dioxide (CO_2) concentrated candle jar at 35 degrees centigrade for 24hrs and

observed for bacterial growth. MacConkey and blood agar were incubated aerobically and observed for 24hrs. If no growth was observed, they were further incubated for 48hr. Potential contaminants were identified by their staining characteristics, appearance of colonies and haemolysis features. Other tests including coagulase, catalase, oxidase, urease, indole, lactose fermentation, mannitol fermentation, motility characters and production of H_2S were also used for further identification. All reagents were purchased from Oxoid limited company, UK. To consider a stethoscope diaphragm contaminated, we used a cut-off colony count of 25 CFU/diaphragm.

Antimicrobial susceptibility test

Antimicrobial susceptibility testing was done using a disk diffusion method as per Clinical Laboratory Standards Institute (CLSI 2011) guidelines [18]. Our antibiotic discs (Oxoid Company Limited, UK) were selected based on which antibiotics are most commonly used in the study area. Discs for both grams positive as well as negative bacteria contained the following antibiotics: Trimethoprim-sulphamethoxazole 25ug, Cefoxitin 30ug, Ampicillin 10ug, Ciprofloxacin 10ug and Gentamycin 10ug, Clindamycin 2ug, Erythromycin 15ug, and Vancomycin 30ug. Cefoxitin was used specifically for detection of MRSA in this study.

Data analysis

Data were entered, cleaned and analysed using SPSS version 16.0 computer software. A p-value of <0.05 was taken to be statistically significant.

Results

The basic socio-demographic, clinical and laboratory characteristics of the study Participants

Of the 100 study participants, 56 were male and 44 were female. The youngest participant was 23 years old while the oldest was 59 years old; the median age was 28 years. Sixty percent of stethoscopes were from medical students, 21 from residents, 10 from interns and 3 from registrars. The study population was comprised

of 78 doctors and students from in-patient departments, 14 from outpatient departments, 4 from the ICU and 4 from the operating room. Of the 78 from in-patient departments, 19 were from Paediatrics, 18 were from adult Medicine, 18 were from Surgery and 23 were from Obstetrics and Gynaecology.

Stethoscope storage

IPC guidelines cover storage, disinfection and sterilization of stethoscopes. The main locations for stethoscope storage amongst our study participants were their homes, offices and cars. All of these areas are non-clinical areas, which goes against IPC protocols. About 65% of our participants stored their stethoscopes at home, while 35% used their offices and their cars. There was a significant relationship between area of storage and degree of contamination (**Table 1**).

Table 1: Risk factors associated with the detection of pathogens from the stethoscopes at KCMC referral Hospital from February to April, 2014 in Moshi, Tanzania

Variable	Presence or Absence of bacteria in the studied stethoscope		Crude OR ((95% CI)	P-Value
	Contaminated stethoscopes (%)	Uncontaminated stethoscope (%)		
Gender				
Female	20 (58.8)	36 (54.5)		
Male	14 (41.2)	30 (45.5)	0.8 (0.4 – 1.9)	0.683
Residence				
Off-campus	24 (70.6)	45 (68.2)		
In-campus	10 (29.4)	21 (31.2)	0.9 (0.4 – 2.2)	0.805
Position				
Medical Doctors	14 (41.2)	27 (40.9)		
Medical Students	20 (58.8)	39 (59.1)	1.0 (0.4 – 2.3)	0.979
Speciality				
Non-surgical	20 (58.8)	42 (63.6)		
Surgical	14 (41.2)	24 (36.4)	1.2 (0.5 – 2.9)	0.639
Duty station				
(Outpatient)	11 (29.4)	13 (19.7)		
(In-patient)	23 (67.6)	53 (80.3)	0.5 (0.2 – 1.3)	0.164
Interval of disinfecting stethoscopes				
Within a week	10 (29.4)	19 (28.8)		

1 week – 3 month More than 3 month	3 (8.8) 21 (61.8)	17 (25.8) 30 (45.4)	0.3 (0.1 – 1.4) 1.3 (0.5 – 3.4)	0.139 0.555
Storage of stethoscopes At home In the Office Car	 22 (64 .7) 12 (35.3) 0 (0.)	 55 (83.3) 7 (10.6) 4 (6.1)	 4.3 (1.5 – 12.0) - -	 0.007

KEY: *OR- Odd ratio, CI-Confidence Interval*

Bacterial contamination

Of the 100 stethoscopes sampled, 41 (41%) of the stethoscopes were found to be contaminated (>25CFU/diaphragm) by bacteria. Fifty-six stethoscopes belonged to female participants while the rest belong to males. There was more contamination from stethoscopes owned by female participants compared to male, but it was not statistically significant. From contaminated stethoscope diaphragms, a total of 134 bacterial strains were isolated. The maximum isolation per single diaphragm was four species. There were 50 isolates of Staphylococcus aureus and out of those, 18% were MRSA (**Table 2**). Potential pathogens were isolated on 41.2% of the stethoscopes from surgical departments (general surgery, orthopaedic, obstetrics and gynaecology) and on 58.8% of the stethoscopes from non-surgical departments, though the difference was not statistically significant. Of gram-positive isolates, coagulase negative staphylococci (CoNS) was the most frequent isolate (58%), followed by *S. aureus* (37.31%) and Bacillus species (5.22%). Of the gram-negative isolates, Klebsiella spp was the most common (5.97%), followed Proteus spp (4.48%), E. coli (2.24%) and *P. aeruginosa* (1.50%). All *P. aeruginosa* isolates were resistant to the four most commonly used antibiotics in the study area (Trimethoprim-sulfamethoxazole, Gentamycin, Ampicillin and Ciprofloxacin). Half of the Klebsiella and Proteus isolates were resistant to Gentamycin. All gram negative isolates were susceptible to ciprofloxacin.

Table 2: Bacterial profile isolates from stethoscopes used at KCMC referral Hospital from February- April 2014 in Moshi, Tanzania.

S/No	Isolated bacteria	Total no. (%)

1	CoNS	58 (43.28)
2	S. aureus	50 (37.31)
3	Bacillus species	7 (5.22)
4	Klebsiella species	8 (5.97)
5	Proteus species	6 (4.48)
6	E. Coli	3 (2.24)
7	P. aeruginosa	2 (1.50)
8	**Total isolates**	**134 (100%)**

Key: *CoNS- coagulase-negative Staphylococci, S. aureus*- Staphylococcus Aureus, E-Coli- Escherichia coli, P. Aeruginosa- *Pseudomonas aeruginosa*

Antimicrobial sensitivity pattern of the isolates

We selected the eight most commonly-used antibiotics in our hospital to test against our isolates. Eighteen percent of the *S. aureus* and 18% of the ConS isolates were methicillin resistant; 15.5% of the *S. aureus* and 7% of the CoNS isolates were resistant to five classes of antibiotics (fluoroquinolones, macrolides, cephalosporins, aminoglycosides and trimethoprim-sulfamethoxazole). Similarly, a number of Klebsiella ssp, Proteus ssp and *E. coli* isolates had resistance to four antibiotic classes. (Table 2)

Disinfection practice

Of the 100 stethoscopes studied, only 10 (10%) of the owners reported that they disinfect their stethoscopes at least once per week. More than 60% of the participants said that they disinfect their stethoscope at least once every three months. However, there was no significant association between frequency of cleaning and type of isolate as well as number of isolates (Table 1). No one responded that they clean their stethoscopes before and after examining each patient.

Discussion

The introduction of non-invasive medical devices such as stethoscope for the management of patients might contribute to the spread of NIs worldwide. Stethoscopes, which touch many patients' skin, can become fomites and thus must be properly cleaned. In our study, a total of 41% of the stethoscopes were contaminated with potentially pathogenic bacteria. This is less than what other studies have shown.[19] Studies conducted in Brazilian and Nigerian teaching hospitals conducted by Shiferaw and Zuliani-Maluf showed contamination rates at Brazil site to be 11% while in Nigeria 90% [19]. Those two study sites differed in many aspects but the African site showed a higher rate of contamination than the South American site. The different between Brazil studies with our study can be partially explained by the presence of very few, less trained medical doctors on IPC. For Nigeria contamination was higher than our current study which might be due to highly congested hospitals. Also the different weather between African and Caribbean regions might explain partially about difference in detected isolates as well as degree of contamination. Africa-Purino reported higher rate of 57% contamination of stethoscopes, when compared to 41% in our current study [20]. None of our study participants adhered to these guidelines with respect to stethoscope storage and over 95% of them did not abide by methods for the proper cleaning of stethoscopes [8, 21]. The different rate of contamination we saw might also have been contributed to other reasons such as type of disinfectants used, frequency of contamination and nature of patient's studies including infectious and non-infectious patients.

In the current study, the mean total bacterial count on contaminated stethoscopes was $1.02X10^4$CFUs/diaphragm. This is higher than that reported in a study by Whittington et al, that included stethoscopes from intensive care units that had a mean of 190.9CFUs/diaphragm [22]. In Whittington study more than 50% of isolates detected were pathogenic [22]. In terms of isolated species, we isolated more gram positive than gram negative bacteria, which is not surprising given that we were culturing skin flora. Of the one hundred and thirty-four bacterial species isolated, a

significant number were pathogenic. This is in line with other studies that have also found high percentages of pathogenic isolates.

Forty-six percent of our studied stethoscopes were contaminated. In one study done in Nigeria, the contamination rate was 79% [8]. Nigerian study has almost twice as much contamination compare to our study, this can be partially explained by congestion, types of disinfectant and even nature of the patients. Some study participants went further by saying they don't disinfect their stethoscope because they don't have disinfectants. At the site of our study we mix both non-infectious like hypertensive (non- infectious) with infectious cases such as staphylococcus boils. In the Nigerian study, the study participants were sampled from infectious diseases wards rather than common non-infectious wards. In the current study, one possible explanation is that most people did not frequently clean their stethoscopes. We didn't ask about the frequency of stethoscope use or the number of patients seen by a single medical doctor; both of these factors could also contribute to different contamination rates [8,23]. Furthermore, more than half of our study participants were storing their stethoscopes at home, which increase the chance of transferring community microbes to the hospital and vice versa.

The most common isolate was *S. aureus* (37.3%). All isolates seen in the current study were different compares to the studies in other medical equipment and hospital environments. One explanation is the different environment settings between stethoscope and other medical equipment like trolleys and weighing scales surfaces [24, 25]. The total colony count seen in our study shows significant colonization on the diaphragm. These high counts are likely facilitated by the failure of doctors and medical students to practice infection control protocol of disinfecting stethoscope every after each patient examination. The documented survival time of most hospital-acquired organisms on the diaphragm surface of the stethoscope pathogen ranges between 2-18hrs, this range encompasses all the patients that will be seen in a work day [26]. *Staphylococcus aureus* is a significant component of human skin flora, and it has been well described as a primary cause of NIs [16, 17]. It is also the

most common pathogen isolated from stethoscopes, regardless of the study area, methodology and sample size employed,

MRSA is a major challenge in many health care facilities in resource rich settings [27.28]. There is very limited information on the prevalence of MRSA in Sub-Saharan Africa and one study in Dar es Salaam Tanzania among under-five children which showed prevalence of MRSA to be 10.5% [33]. The prevalence of MRSA among our isolates was higher (18%) compare to Dar es Salaam study. The reason might be contributed to the fact that, Dar es Salaam study included children who were health while our study was mixture of children and adults. Another explanation is that, under five were healthier compared to our study which included admitted sick patients. Under five study the isolates where taken from nasal swab while our current study isolates were from stethoscopes means from the skin. Other studies have found stethoscopes with varying rates of contamination with MRSA, ranging from of 0-20% [13, 14, 24]. *S. aureus* showed the least resistance to Ciprofloxacin, which is similar to another study from Ethiopia in which Ciprofloxacin susceptibility ranged from 0-8% [29].

The different rate of stethoscope contamination was not statistically significant between doctors and medical student which was 41.2% and 58.8 % respectively, p =0.979). Both doctors and medical students reported a strikingly low rate of stethoscope disinfection, lower than in a study by Uneke et al in Nigeria [8]. There was a difference in level of contamination between groups working in non-surgical units and groups working in surgical units, but the difference was not statistically significant, perhaps due to small sample size. It has been shown that an appropriate supply of disinfecting materials is critical to ensure provider compliance with infection control practices. Absence of disinfection solutions or disinfection points in the hospital likely rank high amongst the reasons for poor compliance with infection control practices amongst doctors and students. Additionally, there is an insufficient focus in the medical training curriculum on the burden of NIs and on strategies to reduce the rates of NIs in daily medical practice. This has been pinpointed by other investigators as a factor contributing to the spread of NIs in northern Tanzania [30-

33]. The majority of the study participants responded that they do not disinfect their stethoscopes frequently for many reasons including the absence of disinfectant, congestion of the wards as well as little encouragement to make regular disinfection a regular habit.

Study Limitations

Our study has several limitations. One was the use of convenience sampling to select study participants. While we did make an effort to include several different clinical departments in our study population, our sampling technique may make our results less generalizable. Also, questionnaire results were prone to recall bias. Given our personal familiarity with our study participants, they may have felt pressure to report better infection control practices than reality. However, despite this, no one reported ideal practices.

Conclusion

The degree of stethoscope contamination found amongst doctors and medical students in our hospital in Tanzania is alarming. Stethoscopes are potentially important vehicles for transmission of NIs to hospitalized patients. Furthermore, the isolated pathogens were resistant to multiple classes of antimicrobial agents commonly prescribed in the study area. Most of interviewed medical doctors and students store their stethoscope in non-clinical areas hence there is a risk of transmitting these pathogens to the community. Attention to issues of poor antibiotic stewardship and poor infection control practices are urgently needed in our setting.

Declaration

Competing interest

The authors declare that they have no competing interest.

Author's contribution

ERS, DWG and **NS** made contributions to the study design and lead data acquisition as well as analysis and interpretation of data. **ERS** drafted the first manuscripts; **EGF**

and **BMN** processed the samples and primary analysis. **ERS, SBK** and **DWG** did analysis of the data and reviewed the first manuscript.

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