

# Investigating potential sources of transmission of healthcare-associated infections in a regional hospital, Ghana

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## ABSTRACT

**Background:** Recent research has shown that healthcare-associated infections (HAIs) are on the increase despite education. **Aims:** The aims of this study were to isolate, quantify, and determine antibiotic susceptibility pattern of bacteria on fomites at the Central Regional Hospital, Cape Coast, Ghana. **Settings and Design:** Purposive sampling of likely areas of contamination and contact by patients and healthcare workers was undertaken. **Materials and Methods:** A total of 100 swabs were taken from door handles, working surfaces, beds and taps from the various wards, consulting rooms, OPDs, laboratory, and surgical theatre. Serial dilution was used in quantifying bacteria, MacConkey and blood agars were used in isolation, and the Kirby Bauer method applied in antibiotic sensitivity testing. Statistical analysis: Data were statistically analyzed using Statview from SAS Version 5.0. The means were separated using double-tailed paired means comparison. **Results:** Mean bacterial count ranges from least in wards ( $9.67 \times 10^{11}$ ), working surfaces ( $1.64 \times 10^{12}$ ), door handles ( $1.71 \times 10^{12}$ ), and highest in taps ( $2.08 \times 10^{12}$ ). Door handles had the highest isolation (23) and highest number of differential isolates were from working surfaces (7). Of the total bacterial isolates, 46.14% were pathogenic, with *S. aureus* being the highest (14.42%), while 53.86% were nonpathogenic made up of 45.2% of *Bacillus spp.* Gentamicin was 100% effective, while 6 of the total 12 antibiotics tested (50%) were 100% resistant in either gram-positive or gram-negative bacteria. **Conclusion:** There was a high potential of bacterial transmission from the studied surfaces requiring hospital management to monitor and enforce cleaning regimen to prevent HAIs.

**Key words:** Antibiotic sensitivity testing, *Bacillus spp.*, healthcare-associated infections, nonpathogenic, pathogenic


## INTRODUCTION

A prevalence survey in 2002 conducted under the auspices of the World Health Organization (WHO) in 55 hospitals of 14 countries representing four WHO Regions (Europe, Eastern Mediterranean, South-East Asia, and Western Pacific)

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showed that an average of 8.7% of hospital patients had healthcare-associated infections (HAIs).<sup>[1]</sup> The human cost is over 99,000 deaths per year in the United States, which represents a 5% death rate for HAIs.<sup>[2]</sup> In March 2009, the Center for Disease Control (CDC) released a report estimating overall annual direct medical costs of HAIs that ranged from \$28 to 45 billion.<sup>[3]</sup> This is particularly important in developing countries where very little amount of resources are available for use for an unbearable number of patients. It is believed that one-third of nosocomial infections are considered preventable and that as many as 92% of deaths from hospital infections could be prevented.<sup>[4]</sup>

It is extrapolated that the rate of incidence of HAI infections in Ghana is approximately 152,000 out of 20.7 million people.<sup>[5]</sup> Bacteria that are often found in the healthcare environment include coagulase-negative *Staphylococcus*, *Bacillus*, *Corynebacterium*, *Streptococcus*, *Clostridium perfringens*, *Enterococcus*, and *Staphylococcus aureus* and gram-negative

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bacteria, many of which can survive on inanimate surfaces for months.<sup>[6]</sup> A research in Ghana isolated a total of 187 (85.8%) bacteria made up of 55.5% nonpathogenic and 30.3% pathogenic organisms from fomites in a regional hospital.<sup>[7]</sup> However, the above study neither quantify nor determine antibiotic susceptibility pattern of bacterial isolates, which was undertaken in the present study in another regional hospital with the aim of accessing levels of contamination as well as the effectiveness of antibiotics against these isolates. This study will further highlight the risks of HAIs in Ghanaian hospitals and call the Health Ministry to action.

## MATERIALS AND METHODS

The study was undertaken at the Regional Hospital, Central Region, Ghana, a referral hospital that serves the people of Cape Coast and the entire Central and satellite areas of the Western Regions in Ghana between September, 2010 and April, 2011.

### Sampling

The organisms were isolated from fomites, of which taps, working tables and benches, main door handles, door surfaces of laboratory, theatre surfaces and folder holders, bed handles, etc., in the different hospital areas that might serve as sources for transmission of HAIs within the hospital were selected based on their contact frequency with both patients and healthcare workers.

A swab stick was used to make swabs of the above areas in consulting rooms, male and female surgical wards, OPDs, pediatrics indoor, phlebotomy room and obstetrics and gynecology area, intensive care unit (ICU), laboratory, and consulting rooms. Samples were taken in a 6-month period, bimonthly, 2 weeks in each sampling month leading to the same surface being sampled thrice. A total of 100 swabs were obtained for the research. Controls of sterile swabs and media were undertaken through all the laboratory procedures at each sampling period to ensure research accuracy.

### Laboratory methods and procedures

#### *Inoculation*

Swabs from sampled surfaces were inoculated in 10 ml of bacteriological peptone water by cutting the swabs aseptically into the peptone water, shaking, and incubating them over-night at 37°C to encourage the growth of especially slow-growing bacteria. This ensures that all organisms contaminating the surfaces were isolated and enumerated.

#### *Quantification of bacteria*

Serial dilutions from the resulting growth from the peptone water medium were pour-plated on count agar and incubated

for 24 h at 37°C under aerobic condition. The number of estimated colony forming units for each sample was then counted using the quebec colony counter (Reichert, USA).

#### *Isolation of organisms*

All pure isolated colonies were subcultured onto blood agar plates (for growth of heterotrophic bacteria) and MacConkey agar plates (for coliforms) for 24 h at 37°C for colony isolation and morphological identification.

#### *Identification of organisms*

Pure isolated colonies were gram differentiated and then biochemically identified using indole, catalase, citrate, oxidase, coagulase, and urease tests.

#### *Antibiotic susceptibility test*

Antibiotic susceptibility were determined by agar diffusion technique on Mueller-Hinton agar (Kirby-Bauer NCCLS modified disc diffusion technique) using eight antibiotics discs (Biotec Lab.UK) corresponding to drugs commonly used in the treatment of human and animal infections caused by bacteria. Antibiotics for gram-negative bacteria include ampicillin (AMP) (10 µg), cefuroxime (CRX) (30 µg), cotrimoxazole (COT) (25 µg), cefotaxime (CTX) (30 µg), tetracycline (TET) (30 µg), amikacin (AMK) (30 µg), gentamicin (GEN) (10 µg), and chloramphenicol (CHL) (30 µg); while for gram-positive bacteria include ampicillin (AMP) (10 µg), cefixime (CXM) (30 µg), cloxacillin (CXC) (5 µg), cotrimoxazole (COT) (25 µg), tetracycline (TET) (30 µg), penicillin (PEN) (10 µg), gentamicin (GEN) (10 µg), and erythromycin (ERY) (15 µg).

#### *Statistical analysis*

Data obtained in the study were descriptively and statistically analyzed using Statview from SAS Version 5.0. The means were separated using double-tailed paired means comparison. A *P* value of < 0.05 was considered significant.

## RESULTS

Mean bacterial counts from the surfaces after swabbing ranged from a least of  $9.67 \times 10^{11}$  from the wards to a highest of  $2.08 \times 10^{12}$  from taps. One-way double-tailed analysis of variance of the different surfaces gave an insignificant value (*P* = 0.06). Bacterial isolates from the various wards was the highest [29, 32.5%] with the least obtained from working surfaces [17, 19.0%] [Table I]. A double-tailed paired means comparison of the surfaces against each other was not significant for the surfaces (*P* > 0.05) except for that of wards against the other surfaces (*P* < 0.05).

*Bacillus spp.* was the highest individual bacterial isolate 63 (45.20%), followed by *S. aureus* 20 (14.42%), with the least

being *Enterobacter spp.* 1 (0.96%) [Table 2]. Pathogenic bacteria isolated from the surfaces were 56 (46.14%), of which *S. aureus* was the highest 20 (14.42%) and *P. aeruginosa* the least 7 (4.80%). *Bacillus spp.* made up for 63 (45.2%) isolates, followed by *Citrobacter spp.* 11 (7.7%) and *Enterobacter spp.*, i.e., 1 (0.96%) of the 75 (53.86%) nonpathogenic isolates. There was no significant difference between pathogenic and nonpathogenic isolates ( $P = 0.592$ ).

Highest number of isolates [14 (25.0%)] were found from laboratories, followed by the male ward and consulting rooms, i.e., 9 (16.1%) each and the least isolates obtained from obstetrics and gynecology and ICU, i.e., 3 (5.4%) each.

Obstetrics and gynecology and ICU had the highest percentage of pathogenic isolates (66.7% each), while the male ward and the consulting rooms had the highest percentage of nonpathogenic isolates (55.6%) [Table 3].

*K. pneumoniae*, *Citrobacter spp.*, *S. pneumoniae*, and *E. coli* were the most resistant organisms showing resistance to 6 (75%) of the antibiotics tested on them. *S. aureus*, *P. aeruginosa*, and *Enterobacter spp.* were resistant to 5 (62.5%) of the antibiotics, while *Bacillus spp.* was the least resistant organism, i.e., 4 (50%) [Table 2]. Of the antibiotics used, gentamicin was the most effective (100%) on both gram-positive and gram-negative organisms. Cotrimoxazole was 100% effective in

**Table 1: Distribution of isolates from major swabbed surfaces\***

Isolates	Working surfaces (%)	Door Handles (%)	Taps (%)	Wards (%)
<i>Bacillus spp.</i>	7 (7.9)	8 (9.0)	9 (10.1)	15 (16.8)
<i>S. aureus</i>	3 (3.4)	3 (3.4)	4 (4.5)	6 (6.7)
<i>K. pneumoniae</i>	2 (2.2)	3 (3.4)	-	3 (3.4)
<i>P. aeruginosa</i>	1 (1.1)	2 (2.2)	2 (2.2)	-
<i>Citrobacter spp.</i>	1 (1.1)	2 (2.2)	2 (2.2)	2 (2.2)
<i>S. pneumoniae</i>	1 (1.1)	3 (3.4)	-	-
<i>E. coli</i>	2 (2.2)	2 (2.2)	2 (2.2)	3 (3.4)
<i>Enterobacter spp.</i>	-	-	1 (1.1)	-
Pathogenic	9 (52.9)	13 (56.5)	8 (40.0)	12 (41.4)
Nonpathogenic	8 (47.1)	10 (43.5)	12 (60.0)	17 (58.6)
Total	17 (19.0)	23 (25.8)	20 (22.3)	29 (32.5)
Mean count ± SD	$1.64 \times 10^{12} \pm 5.25 \times 10^{11}$	$1.71 \times 10^{12} \pm 8.04 \times 10^{11}$	$2.08 \times 10^{12} \pm 2.72 \times 10^{11}$	$9.67 \times 10^{11} \pm 2.97 \times 10^{11}$

\*Data obtained from all swabbed surfaces not shown in this table

**Table 2: Frequency of isolates, percentage resistance, and pattern to antibiotics used**

Isolates	Frequency (%)	Resistance pattern*	Resistance (%) <sup>†</sup>
<i>Bacillus spp.</i>	63 (45.20)	AMP, CXC, CXM, PEN	50.0
<i>S. aureus</i>	20 (14.42)	AMP, PEN, CXC, CXM, TET	62.5
<i>K. pneumoniae</i>	17 (12.50)	AMP, CHL, COT, CRX, CTX, TET	75.0
<i>P. aeruginosa</i>	7 (4.80)	AMP, AMK, CHL, CRX, CTX	62.5
<i>Citrobacter spp.</i>	11 (7.70)	AMP, AMK, COT, CRX, CTX, TET	75.0
<i>S. pneumoniae</i>	8 (5.77)	AMP, PEN, CXC, CXM, ERY, TET	75.0
<i>E. coli</i>	12 (8.65)	AMP, AMK, COT, CRX, CTX, TET	75.0
<i>Enterobacter spp.</i>	1 (0.96)	AMP, CHL, CRX, CTX, TET	62.5

\*Resistance pattern constructed from antibiogram; antibiotic codes as defined in methodology; <sup>†</sup>% resistance calculated from antibiogram

**Table 3: Distribution of isolates in the wards and other areas with their pathogenicity**

Isolates	Male ward	Female ward	Peds	OBG	ICU	Cons room	Theat	Labs
<i>Bacillus spp.</i>	5 (8.9)	4 (7.1)	3(5.3)	1 (1.8)	2 (3.6)	5 (8.9)	3 (5.3)	4 (7.1)
<i>S. aureus</i>	2 (3.6)	1 (1.8)	-	2 (3.6)	1 (1.8)	-	3 (5.3)	3 (5.3)
<i>K. pneumoniae</i>	-	3 (5.3)	-	-	-	3 (5.3)	-	2 (3.6)
<i>P. aeruginosa</i>	-	-	-	-	-	-	-	1 (1.8)
<i>Citrobacter spp.</i>	-	-	-	-	-	-	-	2 (3.6)
<i>S. pneumoniae</i>	-	-	-	-	-	1 (1.8)	-	1 (1.8)
<i>E. coli</i>	2 (3.6)	-	1(1.8)	-	-	-	-	1 (1.8)
<i>Enterobacter spp.</i>	-	-	-	-	-	-	-	-
Pathogenic	4 (44.4)	4 (50.0)	1(25.0)	2 (66.7)	2 (66.7)	4 (44.4)	3 (50.0)	8 (57.1)
Non-Pathogenic	5 (55.6)	4 (50.0)	3(75.0)	1 (33.3)	1 (33.3)	5 (55.6)	3 (50.0)	6 (42.9)
Total	9 (16.1)	8 (14.2)	4(7.1)	3 (5.4)	3 (5.4)	9 (16.1)	6 (10.7)	14 (25.0)

Peds: Pediatrics; OBG: Obstetrics and Gynecology; Cons: Consulting; Theat: Theatres

**Table 4: Effectiveness of antibiotics used in the antibiotic sensitivity test**

Antibiotic (ug/disk)	Gram-positive organisms		Gram-negative organisms	
	Susceptible (%)	Resistant (%)	Susceptible (%)	Resistant (%)
Gentamicin (10 µg)	100	0.00	100	0.00
Cotrimoxazole (25 µg)	100	0.00	50	50
Tetracycline (30 µg)	66.7	33.3	83.33	16.67
Ampicillin (10 µg)	0.00	100	0.00	100
Erythromycin (15 µg)	66.7	33.3	-	-
Cefixime (30 µg)	0.00	100	-	-
Cloxacillin (5 µg)	0.00	100	-	-
Penicillin (10 µg)	0.00	100	-	-
Chloramphenicol (30 µg)	-	-	66.7	33.3
Amikacin (30 µg)	-	-	50	50
Cefuroxime (30 µg)	-	-	0.00	100
Cefotaxime (30 µg)	-	-	0.00	100

gram-positive organism but only 50% so in gram-negative organisms. Tetracycline had varying effectiveness of 66.7% in gram positive and 88.3% in gram negative. Ampicillin was ineffective (100%) in both gram-positive and gram-negative organisms. 6 of the 12 antibiotics tested were resistant either in gram-positive or in gram-negative organisms [Table 4].

## DISCUSSION

Results from the study showed a very high mean range of bacteria count of  $9.67 \times 10^{11}$  to  $2.08 \times 10^{12}$ . This high bacterial count can serve as effective inoculums resulting in HAIs especially in an immune-compromised environment such as a hospital. One-way double-tailed ANOVA of the different surfaces gave an insignificant value ( $P = 0.06$ ). This suggests that all the surfaces have equal potential of causing HAIs. Pathogenic isolates made up 46.14% of the isolated bacteria, while nonpathogenic bacteria were 53.86% with an insignificant difference between them ( $P = 0.592$ ). This indicates a very high pathogenic environment with a high risk of HAIs. This is because pathogenic organisms have high virulence with the potential to cause infections in even healthy individuals, thus placing both healthcare workers and patients at risk of infections regardless of their health status. This finding conforms to a research undertaken in another Regional Hospital in Ghana, though bacterial isolates was higher in this study.<sup>[7]</sup> The reason for this high bacterial isolation could be due to infrequent cleaning in the hospital due to lack of water since the region has a dire water problem. Previous studies showed a low prevalence of pathogenic isolates (15%) in a hospital ward, resulting in a 10% chance of patients acquiring HAIs.<sup>[4]</sup>

The high presence of *S. aureus*, 20 (14.42%), *E. coli*, 12 (8.65%), *K. pneumoniae*, 17 (12.50%), and *P. aeruginosa*, 7 (4.80%), conforms to the earlier researches.<sup>[8,9]</sup> *Bacillus spp.* was the highest nonpathogenic isolate due to its wide distribution and

colonization of several surfaces. This confirms the ubiquitous nature of the organism giving it greater colonization ability as well as the ability of its spores to resist environmental changes, withstand dry heat, and certain chemical disinfectants for moderate periods.<sup>[9]</sup> The wards had the highest isolates, 29 (32.5%), followed by door handles, 23 (25.8%), taps, 20 (22.3%), and working surfaces, 17 (19.0%). The reason for this trend could be due to the fact that working surfaces are visible, and thus, there is a tendency to attempt cleaning, surfaces in the wards such as handles on beds, folder holders on patients beds, drug trays, etc., tend to see little cleaning although these surfaces experience much handling leading to a high level of contamination. The level of isolation of *S. aureus* and the level of pathogenic isolates 8 (40.0%) on taps confirm earlier works that found taps to usually have the most pathogenic isolates but the least total bacterial colonization as a result of their persistent contact with detergents during washing and thus would only be colonized by bacteria that are resistant to the detergent or one that was left there just before swabbing was done.<sup>[10]</sup>

Though total bacterial isolates from the theatre was not the highest 6 (10.7%), percentage of pathogenic isolate (50%) from the theatre was high for a place where several invasive procedures are performed. Researchers have shown that the incidence of HAIs in the theatre during surgery varies from 0.5% to 15% and is dependent on the type of operation and underlying patient status.<sup>[11]</sup>

The results on antibiotic susceptibility testing showed varied degrees of antibiotic resistances among the isolates. Isolates showed 100% resistance to ampicillin, penicillin, and cefuroxime, confirming similar result observed in bacterial isolates found in sachet water sold in the streets of Cape Coast.<sup>[12]</sup> This indicates spread of resistant isolates from hospital to the community and vice versa. This has major health implication in times of epidemics since these antibiotics are the first-line

drugs prescribed by healthcare workers and used by patients during infections. Marked bacterial resistance to ampicillin has been observed in earlier research too.<sup>[13]</sup> Most of the bacterial isolated were resistant to commonly used antibiotics such as ampicillin, penicillin, cefuroxime and cloxacillin (100%), and amikacin (50%) and posed a public health concern in an earlier study.<sup>[14]</sup> Furthermore, majority of the pathogenic isolates 80% showed up to 75% resistance to the antibiotics used indicating that in situation of such infections, these antibiotics may not work. Gentamicin was 100% effective in both gram-negative and gram-positive organisms, which confirms earlier research on *E. coli* isolated from drinking water and milk.<sup>[15,16]</sup> A study in Cape Coast on antibiotic use showed that 66.9% of the populace sampled purchases antibiotics in the open market without any prescription.<sup>[17]</sup> This could be as a result of an influx of antibiotics into the Ghanaian market from both foreign and local pharmaceutical companies, which has led to the considerable increase in the abuse of antibiotics as a result of the Over-The-Counter services without prescription from qualified physicians.<sup>[18]</sup> This trend has been documented to favor antibiotic abuse.<sup>[19]</sup>

All sampled areas of the hospital had a greater than 15% pathogenic isolates of which majority were 75% resistant to commonly used antibiotics, some of which were 0% effective on the isolated bacteria. This study has demonstrated a high possibility of transmission of HAIs due to high levels of contamination of several surfaces by both pathogenic and nonpathogenic bacteria with high resistances to commonly used antibiotics.

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