Rethinking Contextual Waste Management Policies to Mitigate the Risk of Antimicrobial Resistant Infections in Healthcare Settings: Evidence from Selected Hospitals in West Cameroon

**ABSTRACT**

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| **Background:** In large numbers of low- and middle-income countries, hospital waste management policies are conducted below the minimum standards due to resource scarcity. In these areas, accumulated wastes often represent potent sources of microbial populations that may evolve as health threats to humans, animal’s and environmental health, further exacerbating poverty. Hospital wastes globally refer to the waste generated by activities in healthcare facilities. They are produced in patient care units, medical biology laboratories, medical imaging departments, hospital pharmacies, laundry premises, catering and administration units. From these origins, 15% of the waste is hazardous (often toxic and lethal). This issue highlighted the need for redefining, reorienting and optimising waste management policies in these and other healthcare facilities in West Cameroon in order to ensure a better biosafety/biosecurity tandem for hospital users, in line with the above three SDGs concerned with poverty alleviation, healthcare provision and quality education, respectively. **Objective:** The aim of this cross-sectional study was to investigate the type, the diversity, the load and the drug susceptibility trends of bacteria populations that grow in the vicinity of solid waste accumulation sites in four healthcare facilities of West Cameroon. **Methods:** This cross-sectional study was conducted from January 10th through May 15th, 2024, in four healthcare facilities in the West region of Cameroon. Specimen collection was performed in the vicinity of solid waste accumulation sites at the “Université des Montagnes” Teaching Hospital (UdMTH), the Bangangté District Hospital (BangDH), the Bangwa Protestant Hospital (BPH) and the Bandjoun District Hospital (BandDH). Laboratory screening of the specimens was carried out at the UdMTH Laboratory of Microbiology. Soil and air specimens were collected for bacterial screening at varying distances from the solid waste accumulation sites. Culture, isolation, identification, enumeration and susceptibility tests on bacterial isolates were performed according to standard protocols. *Staphylococcus aureus* ATCC 29213 and *Escherichia coli* ATCC 25922 were used as reference bacterial strains for quality control throughout the process. **Results:** Relevant findings revealed diversified populations made up of *Staphylococcus* spp. (50.94%), Gram-positive rods (26.42%), *Acinetobacter* spp. (11.32%), *Klebsiella* spp. (7.55%), *Serratia* spp. and *Pseudomonas* spp. (1.89% each). In terms of their loads, Gram-negative rods loads were higher than those observed with other morphological types. Further details indicated higher loads and diversities in soil specimens collected in the vicinity of solid waste accumulation sites. These trends in loads and diversity were particularly obvious in one of the four target healthcare institutions. The susceptibility tests revealed multidrug resistance, with the highest rates recorded against beta-lactam antibiotics. The most effective drugs consisted of Gentamicin, Clindamycin, Erythromycin, and Trimethoprim/Sulfamethoxazole. **Conclusion:** These findings are indications that exposed human populations are at risk of contracting resistant bacteria, with a higher likelihood in the vicinity of accumulated wastes in all settings. Accordingly, contextual implementation of hospital hygiene policies in line with biosafety and biosecurity was suggested as a priority to meet the expectations of the 1st, 3rd and 4th United Nations Sustainable Development Goals. |

*Keywords:* Bacterial diversity, Resistance, Waste accumulation sites, Hospital solid wastes, Health

1. INTRODUCTION

Hospital wastes globally refer to the waste generated by activities in healthcare facilities. They are produced in patient care units, medical biology laboratories, medical imaging departments, hospital pharmacies, laundry premises, catering and administration units. From these origins, 15% of the waste is hazardous (often toxic and lethal). Hazardous wastes consist of sharp, infectious, pathological, pharmaceutical, cytotoxic, chemical and radioactive pollutants [1,2]. Hospital waste management is an important process that must be dealt with diligently. The management of hazardous waste material requires specific knowledge and regulations, and it must be carried out by specialists in the field (Arshad et al.,2011; Al-Momani et al.,2019).

Similar to any waste management, appropriate hospital waste handling practices should include segregation, containment, internal collection, storage, transportation (external collection), treatment (particularly for healthcare risk waste), and final disposal (Chaerul et al.,2008; Ranjbari et al.,2022). Hazardous waste is defined as waste that poses a significant or potential threat to the environment as well as human health. Hazardous waste handling may be quite difficult. Even on a small scale, improper hazardous waste management can threaten both human health and the environment (Goel & Sharma,2022). In large numbers of low- and middle-income countries’ hospitals, hazardous waste items that are not often separated from the non-hazardous ones cause functional gaps which conflict with standard waste management procedures [1-4]. In some instances, these wastes are stored and eventually treated in the vicinity of the patient’s caretaking premises [2,5,6]. When post-accumulation treatments are poorly conducted, all derivatives become serious threats to human, animal and environmental health [7-9]. In fact, the accumulated wastes represent risk factors for the build-up of toxic and recalcitrant chemical compounds in soils beneath and around their accumulation sites, likely to disrupt the local ecological system equilibrium [10-12].

Moreover, these accumulated wastes represent potent reservoirs for professional and opportunistic pathogenic microorganisms that could interact with their human hosts and cause range of damages with regard to their virulence and the exposed host defence potentials or vulnerability. They are also regarded as sources for selection and dissemination of antibiotic resistance phenotypes and genotypes in local bacterial populations that may spread into the surrounding human communities [13-16]. With the risk of environmental spread and according to certain authors, the risk of hospital-acquired resistant infections is high in both indoor patients and amongst people in communities [17,18].

Managing this risk and preventing infections are integral parts of the global hospital hygiene endeavours that aim at meeting the challenges of the 1st, 3rd and 4th Sustainable Development Goals (SDGs) by 2030. Any management and prevention initiatives could only be carried out effectively if the units in charge of hospital hygiene have related relevant information. In this frame, the aim of the present study was to provide pieces of information (type, load and drug susceptibility) concerning the bacterial populations that are present in the environment of solid waste accumulation sites within four healthcare institutions in the West region of Cameroon. More specifically, this investigation aimed at identifying and quantifying potential harmful bacteria from soil and ambient air in the solid waste accumulation sites, and addressing isolates' susceptibility to common conventional antibacterial agents. Upon completions, overall findings highlighted the need for redefining, reorienting and optimizing waste management policies in these and other healthcare facilities in West Cameroon to ensure a better biosafety/biosecurity tandem for hospital users, in line with the above three SDGs concerned with poverty alleviation, healthcare provision and quality education, respectively. These goals are critical with the increased global life expectancy and projected related healthcare challenges like resistant opportunistic infections, and the overall human welfare.

2. Materials and Methods

**2.1 Study design and ethical/administrative considerations**

This cross-sectional study was conducted from January 10th through May 15th, 2024, in four healthcare facilities in the West region of Cameroon. Specimen collection was performed in the vicinity of solid waste accumulation sites at the “Université des Montagnes” Teaching Hospital (UdMTH), the Bangangté District Hospital (BangDH), the Bangwa Protestant Hospital (BPH) and the Bandjoun District Hospital (BandDH). Laboratory screening of the specimens was carried out at the UdMTH Laboratory of Microbiology.

**2.2 Sample collection**

**2.2.1 Solid waste accumulation sites**

For investigation purposes, all samplings were carried out around accessible and used solid waste accumulation sites (SWAS). These SWAS included the pits, the incinerators and the temporary storage sites of infectious solid wastes.

**2.2.2 Sampling**

The specimens (surface soil and ambient air around the SWAS) were collected according to Kom Fotso *et al*. [19]. Briefly, about 50 g of surface soil was collected aseptically with a sterile spatula at 1 meter (sampling location A) and 30 meters (sampling location B) from the SWAS, then transferred into sterile pots.

In parallel, airborne bacteria were trapped by passive contact (direct contact with the circulating ambient air) on uncovered Petri dishes containing Mannitol Salt, Cetremide and MacConkey agars provided by Liofilchem®. These culture media were chosen for their role in the selective growth of prominent healthcare-associated infections due to bacterial etiologies. These culture media were exposed for 30 min at 1 meter (sampling location A) and 30 meters (sampling location B) from each SWAS.

After collections, soil samples and exposed culture media in Petri dishes (for airborne bacteria) were immediately conveyed to the laboratory in refrigerated containers (4-8°C) for microbial identification and susceptibility testing according to standard procedures.

**2.3 Sample analysis**

Previous and standard protocols [19,20] were used during this step. *Staphylococcus aureus* ATCC 29213 and *Escherichia coli* ATCC 25922 were used as reference bacterial strains for quality control throughout the process.

At the laboratory, the culture of soil specimens was performed according to the Kom Fotso *et al*. workflows [19] on appropriate culture media. For airborne bacteria, previously exposed agar plates were immediately incubated. Subsequent to bacterial growth after 24 h incubation at 37°C, macroscopic examination and enumeration were followed according to the same workflows [19] with soil surface specimens. With airborne bacteria, however, slight modifications were observed in expressing their loads. More precisely, in this investigation, airborne bacterial loads were expressed as colony-forming unit (CFU)/60 mm diameter Petri dish/30 min.

Thereafter, microscopy characterisation (Gram stain) and biochemical identification tests followed. The Catalase test was used for Gram-positive cocci. The tests for oxidase, carbohydrates (mannitol, lactose, glucose) fermentation, motility, urea hydrolysis, indole and tryptophanase production, and citrate metabolism tests were used for Gram-negative rods. The identification of Gram-positive rods was limited to macroscopy and microscopy.

For all bacteria, a pure subculture was conducted at 37°C for 24 h on nutrient agar for susceptibility tests.

**2.4 Antibiotic susceptibility test**

This step was carried out according to the 2023 recommendations of the “Comité de l’Antibiogramme de la Société Française de Microbiologie” (CASFM) [21]. For the clinical categorisation of GPR with Penicillin G (10 U) and Ceftazidime (30 µg) testing, the 2013 recommendation of CASFM was observed [22]. A total of 16 antibacterial agents were then used on 24 h-fresh colonies grown on nutrient agar. Namely, they were Penicillin G (10 U), Oxacillin (1 µg), Amoxicillin (20 μg) (Amoxicillin (25 μg) for GPR), Amoxicillin/Clavulanic Acid (20/10 μg), Ticarcillin (75 μg), Cefoxitin (30 μg), Ceftazidime (30 µg), Ceftriaxone (30 μg), Imipenem (10 μg), Aztreonam (30 μg), Gentamicin (10 μg), Clindamycin (2 μg), Erythromycin (15 μg), Levofloxacin (5 μg), Tetracycline (30 μg), and Trimethoprim/sulfamethoxazole (1.25/23.75 μg).

**2.5 Data analysis**

The target variables were the diversity of bacteria types, their loads and the associated clinical categories (,susceptible-susceptible at high dose-resistant). Data were recorded and processed with tools from Microsoft Excel 2016. Clinical categories are presented as frequencies in the present paper.

To elude institutions’ identity, letters “W”,” X”, “Y” and “Z” were used in the result and discussion sections to refer to target healthcare institutions.

3. results

**3.1 Bacterial diversities and loads**

From the specimens collected in the SWAS areas, 53 bacterial groups were recovered. Out of these, Gram-positive bacteria literally overwhelmed the isolation rates over Gram-negative. In decreasing rates, they were *Staphylococcus* spp. (50.94%), Gram-positive rods (26.41%), *Acinetobacter* spp. (11.32%), *Klebsiella* spp. (7.55%), *Serratia* spp. and *Pseudomonas* spp. (1.89% each).

In further details, bacterial diversities and loads (Table 1) were highest around the ‘Y’ SWAS, with the overall diversities and bacterial loads highest in all soil specimens. Also, invariably, the highest diversities and bacterial loads were observed near SWAS in all settings. *Staphylococcus* dominated the diversity trends, while the highest bacterial loads were recorded with

**3.2 Bacteria susceptibility profile**

Susceptibility testing carried out on isolates revealed multidrug resistance (Table 2), with the highest rates observed for beta-lactam antibiotics. Gentamicin and the Trimethoprim/Sulfamethoxazole combination proved most effective in general, but for the tests on Gram-positive bacteria, Clindamycin and Erythromycin were added to the list of these effective agents.

**Table 1: Bacterial diversity and loads in the subjected specimens**

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Bacteria categories** | **Solid waste accumulation sites** | **Sampling locations** | **W** | | | **Z** | | | **X** | | | **Y** | | | | | |
| **Gram-positive rods** | ***Klebsiella* spp.** | ***Staphylococcus* spp.** | ***Acinetobacter* spp.** | **Gram-positive rods** | ***staphylococcus* spp.** | ***Acinetobacter* spp.** | **Gram-positive rods** | ***staphylococcus* spp.** | ***Acinetobacter* spp.** | **Gram-positive rods** | ***Klebsiella* spp.** | ***Pseudomonas* spp.** | ***Serratia* spp.** | ***Staphylococcus* spp.** |
| **Airborne bacteria**  **(CFU/60 mm diameter Petri dish/30 min)** | **Pit** | **A** |  | | | - | - | 1110\* | - | 660 | 3600 | - | 1380 | - | - | - | 3000 |
| **B** |  | | | - | - | 720\* | - | - | 2100\* | - | 60 | - | - | - | 240 |
| **Incinerator** | **A** | - | - | 1800 |  | | |  | | | - | - | 15360 | - | - | 2160\* |
| **B** | - | - | 600 | - | - | - | - | - | 1380 |
| **ISW temporary storage site** | **A** | - | - | 120 |  | | |  | | |  | | | | | |
| **B** | - | - | - |
| **Soil bacteria**  **(CFU/g of surface sol)** | **Pit** | **A** |  | | | 6420 | 660\* | - | 15180 | 2730\* | - | 16800 | 1860\* | - | - | - | 2520 |
| **B** | 6000 | - | 2460 | 12000 | 2100 | 1200 | - | - | 12000 | - | - | 1200\* |
| **Incinerator** | **A** | - | 1320 | 2220\* |  | | |  | | | 15360 | - | - | - | - | 5970\* |
| **B** | - | - | 240 | - | 4050\* | - | 3660 | 12000 | - |
| **ISW temporary storage site** | **A** | 4230\* | 9000 | 6240 |  | | |  | | |  | | | | | |
| **B** | - | - | 420 |

ISW: Infectious solid wastes; A: Sampling location 1 meter from the waste accumulation site; B: Sampling location 30 meters from the waste accumulation site

\* Bacterial loads from CFUs belonging to two distinct colony morphotype (macroscopy)

**Table 2.a: Bacterial susceptibility profile**

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Antibiotics** | **W** | | | | | | | | | **Z** | | | | | | | | |
| **GNR** | | | **GPR** | | | ***Staphylococcus* spp.** | | | **GNR** | | | **GPR** | | | ***Staphylococcus* spp.** | | |
| **R** | **SHD** | **S** | **R** | **SHD** | **S** | **R** | **SHD** | **S** | **R** | **SHD** | **S** | **R** | **SHD** | **S** | **R** | **SHD** | **S** |
| **Amox./clav. (20/10 μg)** | 100 | 0 | 0 | 100 | 0 | 0 | - | - | - | - | - | - | 100 | 0 | 0 | - | - | - |
| **Amoxicillin (20 μg)\*** | 100 | 0 | 0 | 0 | 0 | 100 | - | - | - | - | - | - | 0 | 0 | 100 | - | - | - |
| **Aztreonam (30 μg)** | 100 | 0 | 0 | - | - | - | - | - | - | 100 | 0 | 0 | - | - | - | - | - | - |
| **Cefoxitin (30 μg)** | 0 | 0 | 100 | 100 | 0 | 0 | 100 | 0 | 0 | - | - | - | 100 | 0 | 0 | 67 | 0 | 33 |
| **Ceftriaxone (30 μg)** | 50 | 50 | 0 | - | - | - | - | - | - | 50 | 0 | 50 | - | - | - | - | - | - |
| **Ceftriaxone (30 μg)** | 50 | 0 | 50 | 100 | 0 | 0 | - | - | - | 50 | 50 | 0 | 100 | 0 | 0 | - | - | - |
| **Clindamycin (2 μg)** | - | - | - | 0 | 0 | 100 | 57 | 0 | 43 | - | - | - | 0 | 0 | 100 | 33 | 0 | 67 |
| **Erythromycin (15 μg)** | - | - | - | 0 | 0 | 100 | 14 | 0 | 86 | - | - | - | 0 | 0 | 100 | 33 | 0 | 67 |
| **Gentamicin (10 μg)** | 0 | 0 | 100 | 0 | 0 | 100 | 0 | 0 | 100 | 0 | 0 | 100 | 0 | 0 | 100 | 33 | 0 | 67 |
| **Imipenem (10 μg)** | - | - | - | - | - | - | - | - | - | 100 | 0 | 0 | - | - | - | - | - | - |
| **Levofloxacin (5 μg)** | 0 | 0 | 100 | 0 | 0 | 100 | 0 | 100 | 0 | 0 | 0 | 100 | 0 | 0 | 100 | 33 | 67 | 0 |
| **Oxacillin (1 µg)** | - | - | - | 100 | 0 | 0 | 100 | 0 | 0 | - | - | - | 100 | 0 | 0 | 100 | 0 | 0 |
| **Penicillin G (10 U)** | - | - | - | 100 | 0 | 0 | 100 | 0 | 0 | - | - | - | 0 | 100 | 0 | 100 | 0 | 0 |
| **Tetracycline (30 μg)** | 100 | 0 | 0 | 0 | 0 | 100 | 14 | 0 | 86 | - | - | - | 0 | 0 | 100 | 0 | 0 | 100 |
| **Ticarcillin (75 μg)** | - | - | - | - | - | - | - | - | - | 50 | 0 | 50 | - | - | - | - | - | - |
| **Trim./Sulf. (1.25/23.75 μg)** | 50 | 0 | 50 | 0 | 0 | 100 | 14 | 0 | 86 | - | - | - | 0 | 0 | 100 | 33 | 33 | 33 |

GNR: Gram negative rods; GPR: Gram positive rods; - : not tested ;

R : rate of resistance isolate; SHD : rate of isolate susceptible at high dose; S : rate of susceptible isolate;

Amox./clav.: Amoxicillin/Clavulanic Acid (20/10 μg); Trim./Sulf.: Trimethoprim/sulfamethoxazole (1.25/23.75 μg)

\* Amoxicillin (25 μg) for GPR

**Table 2.b: Bacterial susceptibility profile (Contd.)**

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Antibiotics** | **X** | | | | | | | | | | **Y** | | | | | | | | |
| **GNR** | | | **GPR** | | | ***Staphylococcus* spp.** | | | **GNR** | | | | **GPR** | | | ***Staphylococcus* spp.** | | |
| **R** | **SHD** | **S** | **R** | **SHD** | **S** | **R** | **SHD** | **S** | **R** | | **SHD** | **S** | **R** | **SHD** | **S** | **R** | **SHD** | **S** |
| **Amox./clav. (20/10 μg)** | - | - | - | 50 | 50 | 0 | - | - | - | 100 | | 0 | 0 | 50 | 50 | 0 | - | - | - |
| **Amoxicillin (20 μg)\*** | - | - | - | 0 | 50 | 50 | - | - | - | 100 | | 0 | 0 | 50 | 50 | 0 | - | - | - |
| **Aztreonam (30 μg)** | 100 | 0 | 0 | - | - | - | - | - | - | 50 | | 17 | 33 | - | - | - | - | - | - |
| **Cefoxitin (30 μg)** | - | - | - | 50 | 50 | 0 | 100 | 0 | 0 | 33 | | 0 | 67 | 100 | 0 | 0 | 100 | 0 | 0 |
| **Ceftriaxone (30 μg)** | 100 | 0 | 0 | 0 | 0 | 0 | - | - | - | 100 | | 0 | 0 | - | - | - | - | - | - |
| **Ceftriaxone (30 μg)** | 0 | 100 | 0 | 100 | 0 | 0 | - | - | - | 80 | | 0 | 20 | 100 | 0 | 0 | 0 | 0 | 0 |
| **Clindamycin (2 μg)** | - | - | - | 0 | 0 | 100 | 0 | 0 | 100 | - | | - | - | 0 | 0 | 100 | 0 | 0 | 100 |
| **Erythromycin (15 μg)** | - | - | - | 0 | 0 | 100 | 0 | 0 | 100 | - | | - | - | 0 | 0 | 100 | 0 | 0 | 100 |
| **Gentamicin (10 μg)** | 0 | 0 | 100 | 0 | 0 | 100 | 0 | 0 | 100 | 0 | | 0 | 100 | 0 | 0 | 100 | 14 | 0 | 86 |
| **Imipenem (10 μg)** | 100 | 0 | 0 | - | - | - | - | - | - | 100 | | 0 | 0 | - | - | - | - | - | - |
| **Levofloxacin (5 μg)** | 0 | 0 | 100 | 0 | 0 | 100 | 0 | 100 | 0 | 0 | | 17 | 83 | 0 | 0 | 100 | 0 | 100 | 0 |
| **Oxacillin (1 µg)** | - | - | - | 100 | 0 | 0 | 100 | 0 | 0 | - | | - | - | 100 | 0 | 0 | 100 | 0 | 0 |
| **Penicillin G (10 U)** | - | - | - | 100 | 0 | 0 | 100 | 0 | 0 | - | | - | - | 50 | 0 | 50 | 100 | 0 | 0 |
| **Tetracycline (30 μg)** | - | - | - | 0 | 0 | 100 | 50 | 0 | 50 | 25 | | 25 | 50 | 0 | 0 | 100 | 29 | 0 | 71 |
| **Ticarcillin (75 μg)** | 0 | 0 | 100 | - | - | - | - | - | - | 0 | | 67 | 33 | - | - | - | - | - | - |
| **Trim./Sulf. (1.25/23.75 μg)** | - | - | - | 0 | 0 | 100 | 0 | 0 | 100 | 0 | | 0 | 100 | 0 | 0 | 100 | 57 | 14 | 29 |

GNR: Gram negative rods; GPR: Gram positive rods; - : not tested ;

R : rate of resistance isolate; SHD : rate of isolate susceptible at high dose; S : rate of susceptible isolate;

Amox./clav.: Amoxicillin/Clavulanic Acid (20/10 μg); Trim./Sulf.: Trimethoprim/sulfamethoxazole (1.25/23.75 μg)

\* Amoxicillin (25 μg) for GPR

4. discussion

Hospital waste environments are reservoirs of microbes and genes transfer-enabling environments in which cross- and co-selection of antibiotic resistance are predictable events [13-18]. Foremost exposed to these environmental adulterations, the ecological systems within and surrounding these areas are risky for human life, especially for those with compromised immune systems like many in healthcare institutions. The present study, conducted in four healthcare institutions of West Cameroon, revealed that surface soil and airborne bacteria around SWAS were highly diversified, overwhelmed, however, by *Staphylococcus* that represented half of the diversity rates recorded, while Gram-positive and Gram-negative rods each accounted for almost a quarter. This dominance of *Staphylococcus* previously reported in the soil and air around the UdMTH solid waste accumulation sites [19] is most likely in connection with the bacterial cell organization and the non-stringency feature of members from the *Staphylococcus* genus [19,23], consistent with their role as relevant group of bacteria that could effectively be used in hospital hygiene assessment [19,24-26].

In contrast to bacterial diversity, GNR loads were found to be higher than those of other bacterial types. This could be justified by the fact that soil surfaces are richer in easily degradable nutrients (organic substances), which strongly contribute to the residual fitness and perpetuation of Gram-negative bacteria. In addition, Gram-negative bacteria (more copiotrophic) are often dependent on labile carbon supplied by plant litter and other sources, then abundant in surface soils [27-29]. In the context of the present investigation, the SWAS were located in areas covered with vegetation, then humid and conducive for Gram-negative bacteria. Reversely, Gram-positive bacteria (oligotrophic) basically predominate in nutrient-poor environments, beyond the above-mentioned cellular organisation, which allows resistance to environmental stresses like water deficiency. According to previous authors, in fact, bacteria like *Actinomycetes* and other Gram-positive bacteria are common in deeper soils, while Gram-negative populations decrease with increasing depths [27-29]. Otherwise, the type of target specimen may justify the low rates of GPR detection, while deeper soil samples might have provided different values of bacterial diversities and loads. Kom Fotso *et al*. observed that this distribution might also reflect the protocol used, as it was not the most effective one for GPR that are actually expected to grow better on nutrient agar or deMan Rogosa Sharpe agar. Acknowledging therefore that plate count, trypticase soy, nutrient or GPR selective agars could generate different trends, their use will be considered to address this crucial issue during the forthcoming related research initiatives.

Bacterial diversities and loads were lower in ambient air than in soil samples. These findings (soil *versus* ambient air difference) could be attributable to the accumulative and nutritive characteristics of soil compared to the transporter nature (non-accumulative and non-nutritive) of ambient air.

Observing that bacterial diversities and loads were also higher at sampling locations that were closer to the waste accumulation sites basically reflects the presence of conducive requirements like nutrients in the accumulated wastes (that, in turn, serve as microbial reservoirs) and human (hospital staff) activities at distances [16,30] and specifically, from the sampling locations A and B. How each of the related factors (soil contamination by antibiotic resistance genes, antibiotic-resistant bacteria and emerging contaminants; increased nutrient richness, variation in abiotic and biotic entities) directly or indirectly impact variations of bacterial populations as observed by previous authors [16,30,31] is yet to be fully elucidated. Similar future projects are, therefore, expected to provide clearer explanations for the fact that the greatest bacterial diversities and loads were observed in the healthcare institution “Y”. At the same time, however, it was observed that the accumulated waste loads were also bigger at the “Y” where the local vegetation on the SWAS area was most abundantly extended. In line with above arguments, these findings from the “Y” could help anticipate the high environmental contamination from the accumulated wastes, the frequent human activities in the vicinities of waste accumulation sites and the high nutrient richness which in turn corroborates the local high bacterial diversity as well as the anticipable higher likelihood of human affections.

Investigations through the bacterial susceptibility to conventional antibacterial agents revealed high rates of antibiotic-resistant isolates, especially with beta-lactam antibiotics. If these bacteria are generally known to be of hospital origin (from inside hospital premises and spread to the surroundings *via* poor waste disposal or other vehicles), these alarming resistance rates are not surprising. Previous research [25,26,32] reported similar trends on the surfaces and in the air circulating in these hospitals. This resistance trend is at first glance, fundamentally attributable to the selection pressure exerted by antibiotics, antiseptics and disinfectants used in caretaking and hospital hygiene; drug derivatives like heavy metals in wastes; but also, other related selection-driving paths in communities. This involves mobile genetic elements responsible for co-selection and/or cross-selection of resistance phenotypes by the famous traditional and fundamental mechanisms (transduction, conjugation or transformation) that control horizontal genetic transfer within and across bacteria phylogenetic barriers and eventually disseminate in the “hospital – waste – community” frame [14-17;33-37], facilitated by inherent gaps in biosecurity. These phenomena are currently known to be amplified by the use of selection drivers in animal farms and crop production, and encouraged by higher demands that accompany increased human populations and welfare needs. If these bacteria are considered to be of environmental origin and to belong to the SWAS area, these resistances might involve the acquisition of mobile genetic elements spread from accumulated wastes [14,16], a co-selection during the development of tolerance to biocides [37] in the accumulated wastes and to those used during routine management of the SWAS areas, a co-selection during the tolerance process against heavy metals present in the accumulated wastes [14,16,37] or other stressing factors.

Mastering their origins and the pathways they follow represents pressing research challenges with the current One Health paradigm that requires holistic contributions to address all health issues [17,33]. These holistic contributions would guide orientations of contextual waste management policies at all locations, and could extend to other healthcare institutions that share similar environmental variables or be adjusted to suit local realities.

Gentamicin was the most effective drug, followed by Clindamycin and Erythromycin in Gram-positive bacteria. These are advisable broad-spectrum alternatives for potential infections acquired in these hospitals. These antibiotics proved to be effective on some bacterial populations recovered from hospital surfaces and from ambient air of three out of the four target institutions (UdMTH, BangDH, BPH) [25,26,32]; as well as in a parallel survey conducted on bacterial population profile in high-risk infectious premises within the same institutions [38].

Relatively high rates of isolates susceptible to Trimethoprim/Sulfamethoxazole were observed. This finding has become uncommon in human or animal medicine, and resurfaces debate orientations towards the environmental origin of these bacteria. In fact, bacteria recovered from hospital environments in previous studies [25,26,32] and in the above parallel investigation [38] revealed high resistance rates with this drug combination. This contrasting figure also deserves further comparative investigations.

In 2021, Kom Fotso *et al.* [19] reported high rates of susceptible isolates from the UdMTH with a similar investigation protocol. The rate variation between 2021 and 2024 could be, at first glance, in line with bacterial population evolution due to weaknesses in hospital hygiene over time or other factors yet to be properly highlighted. Admitting that the isolates are potential infectious disease etiologies (professional or opportunistic), and that their loads are above infectious doses [39], each of these sites would represent a risky place for patients, especially “Y’. Otherwise, and based on the present findings, basic hygiene policies should be rethought to mitigate the current potentially overlooked healthcare-associated infections risks, though most of the resistant bacterial strains likely disseminate from farms [40-45]. Then, resistance dissemination from farms should also deserve similar consideration in the overall policy regarding the control of resistant infections, which firmly relies on all stakeholders’ education. Accordingly, encouraging observance of biosafety and biosecurity rules in these areas of waste accumulation sites and institutions as a whole appears as a priority necessity to meet the 1st, 3rd and 4th Sustainable Development Goals expectations. This improvement could help prevent the selection and the spread of resistant infectious agents, then mitigate infectious disease rates and related drawbacks in exposed vulnerable populations.

5. Conclusion

The present investigation on waste accumulation site bacteria profile revealed that half of it consisted of *Staphylococcus*, while Gram-positive and Gram-negative rods accounted for a quarter each. In terms of bacterial loads, Gram-negative rod loads were greater than those of the other bacterial types. Their diversities and loads were lower in the air than in the soil samples. The highest bacterial diversities and loads were basically observed in the vicinity of solid waste accumulation sites. Investigations through the susceptibility profile revealed high rates of antibiotic-resistant isolates, especially to beta-lactams, while Gentamicin, Clindamycin, Erythromycin and Trimethoprim/Sulfamethoxazole were most effective. Encouraging observance of rules in line with biosafety and biosecurity in contextual hospital hygiene policies was suggested as a priority necessity to meet the United Nations’ 2030 1st, 3rd and 4th Sustainable Development Goals needs.

COMPETING INTERESTS

Authors have declared that they have no known competing financial interests OR non-financial interests OR personal relationships that could have appeared to influence the work reported in this paper.

ETHICAL APPROVAL

Before field work initiation, all ethical and administrative requirements were fulfilled. Namely, they were the ethical clearance N° 2024/091/UdM/PR/CEAQ obtained from the Université des Montagnes Ethics and Quality Assurance Committee, the research authorizations N° 2024/005/CUM/ADMN\_GENE, 022/A/MINSANTE/DRSPO/HDB/BGTE and 2024/227/UdM/PR/DECANAT-ISSS/MED, respectively provided by the UdMTH, the BangDH, and Université des Montagnes (UdM) Higher Institute of Health Sciences. The directors of BandDH and BPH, respectively, also consented with signed and stamped letters validating project implementation within their institutions.

Disclaimer (Artificial intelligence)

Option 1:

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

Option 2:

Author(s) hereby declare that generative AI technologies such as Large Language Models, etc. have been used during the writing or editing of manuscripts. This explanation will include the name, version, model, and source of the generative AI technology and as well as all input prompts provided to the generative AI technology

Details of the AI usage are given below:

1.

2.

3.

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