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A Brief Report of Environmental Contamination with Gram-Negative Bacteria at Front-Line Hospitals in Northern Syria

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Abstract

Daily operations on wounded soldiers are performed at front-line hospitals. Because diagnostic procedures are not easily available during war and antibiotics are given to patients with suspected infection, it is crucial to assess the resistance patterns of pathogens that colonize the environment at hospitals in order to establish routines and guidelines to decrease antibiotic misuse. In this study, we assessed the source of postsurgical infection caused by multiple resistant gram-negative bacteria by performing culture tests in water samples taken from wastewater, a sink in the operation room, and soap at intensive care units from five military hospitals in northern Syria. Although it was not possible to further characterize the bacteria found on the agarose dishes, cultures taken from moist environments at intensive care units grew colonies of gram-negative bacteria resistant to several available antimicrobial agents. In conclusion, we found that colonized bacteria in pipes, wastewater, soap, or bottles containing disinfectants may be sources of nosocomial infection in postoperative patients. Environmental assessment and regular controls are needed to yield valuable information regarding contamination and the susceptibility of bacteria to antimicrobial agents.

Keywords: Gram negative bacteria • Intensive care unit • Nosocomial infection

Introduction

Northern Syrian is currently one of the most war-torn areas worldwide. Injuries caused by conventional weapons expose susceptible patients to infection in intensive care units (ICUs) or hospital wards. Appropriate diagnostic methods are not available at the majority of front-line hospitals, and antibiotics, if available, are not guaranteed to contain the expected amount of the given drug. Bacterial culture testing is not a routine diagnostic procedure and is seldom used. Antibiotic cocktails are administered as prophylaxis or therapy without available guidelines. Patients may survive the injury but succumb to hospital-acquired infections. For physicians without available diagnostic tools or updated guidelines, it is a great challenge to choose the most appropriate empirical strategy for treatment of the patient. This problem has become even more serious owing to the global spread of antimicrobial resistance, which has been intensified by poor hygiene and excessive use of antibiotics in developing countries. Gram-negative bacteria, including the family *Enterobacteriaceae* and non-lactose fermenting bacteria, such as Pseudomonas and Acinetobacter species, are major causes of hospital-acquired infections in ICUs, accounting for most cases of hospital-acquired pneumonia and urinary tract infections and 25%–30% of bloodstream and surgical infections [1]. Pseudomonas aeruginosa is a free-living bacterium often found in natural waters, such as lakes and rivers. This organism is also found in high-nutrient environments, such as wastewater, and may colonize water systems via biofilm formation. *P. aeruginosa* is the leading cause of illness in immunosuppressed individuals [2]. In hospital environments, contaminated sinks are associated with nosocomial transmission [3]. Airborne dissemination also plays a significant role in patient-to patient spread of epidemic strains of *P. aeruginosa*, suggesting that patients colonized with *P. aeruginosa* should be separated from other immunosuppressed patients [4].

Acinetobacter species have emerged as important nosocomial pathogens responsible for increased mortality and morbidity in hospitalized patients. These organisms can persist in the environment for prolonged periods of time, and environmental contamination has been linked to hospital outbreaks. The surrounding environment is frequently contaminated, and surfaces often touched by healthcare workers during routine patient care are commonly contaminated, making them a source of nosocomial spread [5]. *Enterobacteriaceae* are the most common cause of unexpected in-hospital deaths due to healthcare-associated infection [6]. Although infections due to extended-spectrum β-lactamase (ESBL)-producing gram-negative bacteria are associated with considerable morbidity and mortality among hospitalized patients, only 5% of acquired infections appear to be related to patient-to-patient transmission [7]. Thus, environmental contamination may not play a substantial role in the transmission of ESBL-producing pathogens, and enhancing environmental decontamination may be less effective than other interventions for preventing the transmission of ESBL-producing pathogens [8].

Culture results from tube secretion in a young man on a ventilator at the ICU in a front-hospital in northern Syria (Rojava) yielded growth of non-lactose fermenting gram-negative bacteria, and the resistance profile showed complete resistance to antimicrobial agents, including levofloxacin, cefixime, cotrimoxazole, imipenem, cefadroxil, ceftazidime, lincomycin, doxycycline, ciprofloxacin, and vancomycin. This patient initially suffered from a gunshot wound in the arm. Although not published, several other

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similar cases were observed at the ICU in the same center. *P. aeruginosa* or *Acinetobacter baumannii* were suspected bacterial sources of infection.

Despite shortages in documentation systems, insufficient microbiological diagnostic routines, and the lack of availability of molecular biologic assessments for identification of bacteria, it is crucial to identify the source of infection at ICUs in front-line hospitals. Therefore, in this study, we report our observation from cultures collected in the operation room and ICUs from five military hospitals in northern Syria.

Materials and Methods

Setting

The ongoing civil war in northern Syria has resulted in frequent human injuries, and patients are transferred daily to front-hospitals for treatment. This study was carried out at ICUs, including medical, surgical, and trauma ICUs, at military hospitals in five cities (Al-Qamishli, Al-Hasakah, Manbij, Serê Kaniyê, and Kobanî) in northern Syria.

Cleaning and disinfection policies and procedures

Environmental services policies at the hospitals included in this study called for daily in-room housekeeping activities, including cleaning and disinfection of all patient furniture and spot cleaning of floors using a germicidal cleaner. However, at the time of this study, there were no controlled routines to measure compliance with these policies.

Data collection

Environmental samples were obtained from surfaces in each room (door knobs, bedrails, vital sign monitor touch pads, sinks, wastewater, soap, infusion pumps, ventilator surface touch pads, bottom rails of the doors, and floors). At each site, an area of approximately 10 cm2 was sampled using a sterile cotton swab previously moistened with saline. The sterile swabs were dipped in wastewater spots at or outside the ICU.

Microbiological analysis methods

After collection, each of the environmental swabs was cultured on agar plates (Gélose au sang BioMérieux, France) and EMB agar plates (HiMedia Laboratories Pvt. Ltd., India), prepared at the laboratory according to manufacturer instructions and was incubated for 24 h at 37°C. After 24 h, the colonies were evaluated by gram-staining and subculture to MacConkey II agar plates.

Colonies grown on MacConkey agar plates (both lactose fermenting and non-lactose fermenting organisms) were subculture to agar plates (Gélose au sang BioMérieux) and incubated overnight at 37°C. The sensitivity to different antibiotics was assessed by disc diffusion (Aptek Biologicals Ltd.) breakpoint assays (Table 1).

Antimicrobial agents	Abbreviation	S (over)	R (below)
Ceftazidime	CAZ	18	14
Nitrofurantoin	NIT	17	14
Vancomycin	VAN	12	9
Ciprofloxacin	CPR	21	15
Cefotaxime	CTX	23	14
Gentamycin	GEN	15	12
Cefexime	CFM	19	15
Imipenem	IPM	16	13
Clarithromycin	CLM	18	13
Doxycycline	DOX	16	12
Levofloxacin	LEV	17	15
Amikacin	AK	17	14
Amoxi & Clavulan	AMC-AMX	20	13
Cifuroxime	CXM	18	14
Cotrimoxazole	COT	-	-
Lincomycin	LIN	15	9

Table1. Antimicrobial agents used for the antibiogram.

Results

In cultures collected from wastewater in the ICU, green colonies from non-lactose fermenting gram-negative bacteria that were multidrug resistant (*P. aeruginosa*) were found in the agar plates (Figure 1) (Table 2). The resistance profiles of bacteria found in wastewater in the ICU were identical to the culture results from tube secretion of a patient who was present in the ICU at the time of sampling (Figure 2). The gram-negative bacteria found in other places or hospitals in Manbij, Serê Kaniyê, and Kobanî were lactose fermenting multidrug-resistant bacteria *Enterobacteriaceae* (Figure 3).

	VAN	LEV	DOX	CFX	CAZ	AK	GEN	IPM	CPR	AMC
Suction outlet	R	R	R	R	R	S	S	S	S	R
Wastewater in the operation room	R	R	R	R	R	S	S	R	R	R
Sink in the operation room	R	R	R	R	R	R	R	R	R	R
Gasket of the sink in the operation room	R	R	R	R	R	R	R	R	R	R
Soap and sponge	R	R	S	S	S	S	S	S	S	R
Floor in the operation room	R	R	R	R	R	S	R	R	I	R
Waste basket in the operation room	R	S	R	R	R	S	R	S	I	R
Bottom rail and sill of the door in the operation room	R	S	R	R	I	S	R	S	I	R
Bottom rail and sill of door 2 in the operation room	R	R	R	R	R	R	R	R	R	R
Sink of ICU2	R	R	R	R	R	S	R	R	I	R
ICU2 wastewater	R	R	R	R	R	R	R	R	R	R
Corner of ICU1	R	R	R	R	R	R	R	R	R	R
Wastewater at the pool	R	R	R	R	R	S	R	R	R	R
Wastewater outside the pool	R	R	R	R	R	R	R	R	R	R

Table 2. Environmental contamination in the ICU and operation room at the military hospital in Al-Hasakah VAN =vancomycin, LEV=levofloxacin, DOX=doxycycline, CFX=cefotaxime, CAZ=ceftazidime, AK=amikacin, GEN=gentamycin, IPM=imipenem, CPR=ciprofloxacin, AMC=ampicillin+clavulanic acid

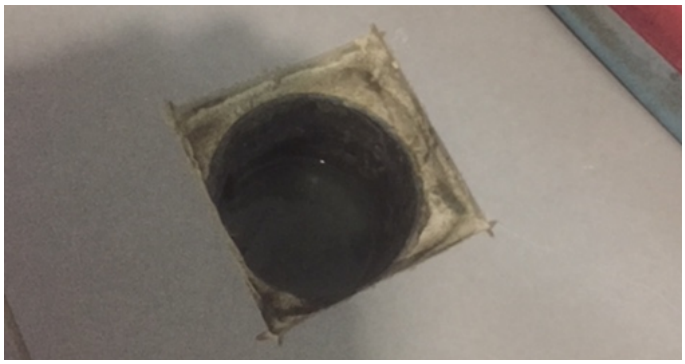


Figure 1. Wastewater at the operation room.

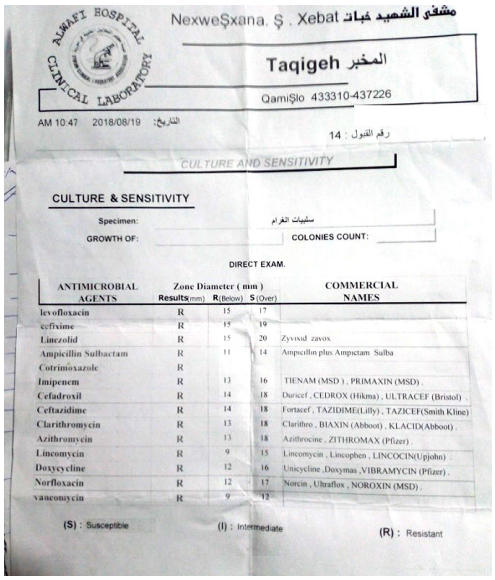
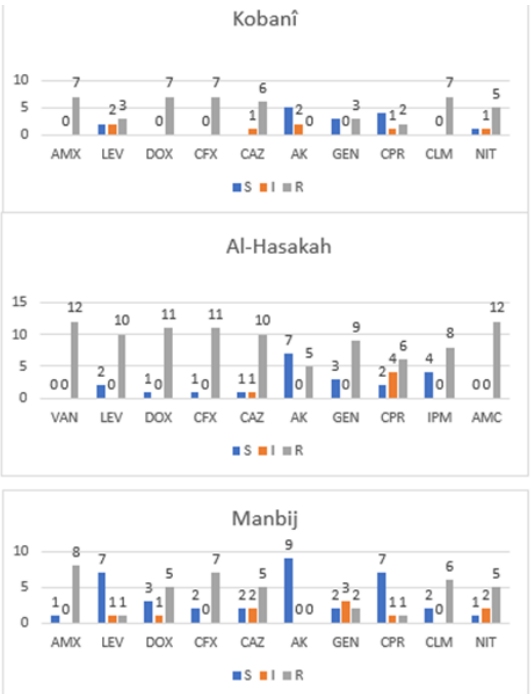


Figure 2. Culture results from tube secretion of a patient suffering from ventilator-associated pneumonia at the ICU.



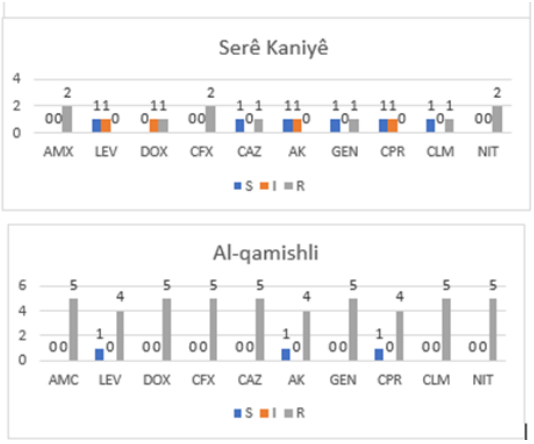


Figure 3. Histograms showing the resistance patterns for gram-negative bacteria found in cultures taken at the ICU and the operation room in military hospitals in Kobanî (n=7), Al-Hasakah (n=12), Manbij (n=9), Serê Kaniyê (n=2), and Al-qamishli (n=5) in northern Syria (Rojava). The military hospital in Manbij was renovated and was not functional.

Discussion

In the current study, we showed that wastewater and bottles of disinfectant solutions at the ICU in hospitals in northern Syria were contaminated with multidrug-resistant gram-negative and non-lactose fermenting bacteria also found in patient cultures at the same ward.

Several studies have highlighted the emerging problem of multidrug-resistant gram-negative bacteria in the Middle East, particularly in Syria [9-13]. However, extensive investigations are still needed to determine the source of the problem and decrease the loss of valuable resources in the affected regions.

Antibiotic misuse is one of the major reasons for the development of multidrug-resistant bacterial strains in the community [14]. The use of counterfeit antimicrobials affects the quality of medical intervention. However, few studies have evaluated the extent of the problem [15,16].

Antimicrobial stewardship (AMS) is the systematic effort to educate and persuade prescribers of antimicrobials to follow evidence-based prescribing in order to decrease antibiotic overuse and thus prevent antimicrobial resistance [17]. AMS programs have been shown to reduce antibiotic use and hospital costs successfully [18-20] and one of the major steps in AMS programs is environmental monitoring [21].

Observation of hospital environments, such as people eating in the corridor outside the ICU, nurses wearing rings and watches under plastic gloves, the use of the same apron when changing wound dressings on patients, lack of isolation facilities, lack of special hospital clothing, cats tearing up waste bags containing wound dressings, and nurses who prescribe antimicrobial cocktails on behalf of doctors to anyone reporting diarrhea, etc., has indicated that there is an urgent need for serious changes in the hospital organizing system. Because Syria has been suffering from civil war for the last 8 years, more loss of life owing to lack of knowledge or discipline should be avoided. However, it may be possible to decrease loss of resources by logical planning and following guidelines. From our results, we found that observations, tests, and documentation were key interventions for identifying the problem, mapping the discrepancies, and convincing decision-makers because foreign guidelines were not suitable in our hospitals.

There were some limitations to this study. Importantly, the research group did not have access to polymerase chain reaction-based microbiological identification methods. Two incubators, an autoclave, light microscopes, empty dishes, agarose powders, dyes, and swabs were used to perform

cultures and study the bacterial strains under microscopes. However, researchers had full access to local citizens and an open environment for discussion, criticism, and involvement. Within 1 week after the report was internally published, the process of decontamination of water sewage by acetic acid solution [3] was initiated at the military hospital in Al-Qamishli.

Conclusion

In conclusion assessment of environmental contamination may facilitate identification of the source of infection and the susceptibility to antibiotics. Management of this problem will save lives and resources. Thus, our findings could have a crucial role in the development of guidelines to limit antibiotic misuse.

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