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The Prevalence and Antimicrobial Resistance of Bacteria Isolated from Surgical Site Infection in Three Large Referral Hospitals in Isfahan, Iran

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Keywords:

Surgical Wound Infection; Microbiology; Microbial Sensitivity Test; Drug Resistance; Iran

Keypoints:

1. E. coli, Staphylococcus aureus and Klebsiella spp. were the most prevalent causes of surgical site infections.
2. Gram-negative isolates showed high susceptibility to Imipenem and Aminoglycosides, and low sensitivity to Third generation Cephalosporins, Ciprofloxacin, and Trimethoprim-Sulfamethoxazole.
3. Gram-positive organisms had high sensitivity to Rifampinand Vancomycin, moderate susceptibility to Aminoglycosides and Trimethoprim-Sulfamethoxazole and low susceptibility to Erythromycin, Clindamycin, Tetracycline and Ciprofloxacin.

1. Abstract

1.1. Aims: Surgical site infection is one of the most prevalent nosocomial infections. Up-to-date knowledge on the etiology and antibiotic susceptibility pattern specific to each geographical region plays a significant role in treating the patients.

1.2. Methods: In a prospective cross-sectional study, the data on the microbiology and antibacterial susceptibility of surgical site infections were collected from March 2016 to March 2018 in three referral hospitals in Isfahan, Iran.

1.3. Results: In total, 90 patients with culture-proven surgical site infection were detected. The most prevalent organism was E. coli (25.5%), followed by Staphylococcus aureus (22.2%), Klebsiella spp. (18.8%), Pseudomonas aeruginosa (8.8%) and Enterococcus spp. (7.77%).

The highest susceptibility of Gram-negative isolates was to Carbapenems (76.5%) and Aminoglycosides (69.8%) followed by Third generation Cephalosporins (36.8%), Trimethoprim-Sulfamethoxazole (27.6%), Ciprofloxacin (25.0%) and Tetracycline (0.0%).

Gram-positive organisms showed high sensitivity to Rifampin (100%), Vancomycin (85.1%), moderate sensitivity to Aminoglycosides (60.8%) and Trimethoprim-Sulfamethoxazole (55.0%), and low susceptibility to Ciprofloxacin (38.5%), Tetracycline (35.0%), Clindamycin (27.3%) and Erythromycin (21.7%).

1.4. Conclusion: our study found that Gram-negative bacilli and Staphylococcus aureus are the most common causes of SSI in understudy hospitals. Moreover, the sensitivity of Gram-negative isolates to imipenem and Aminoglycosides and the susceptibility of Gram-positive isolates to Vancomycin and Rifampin were high, making these drugs the most appropriate antibiotics in treating such infections. Surveillance of antimicrobial resistance and rational use of antibiotics is suggested for the effective treatment of the infection as well as the prevention of microbial resistance in all hospitals.

2. Introduction

Surgical site infection (SSI) is the second most common hospital-acquired infection in Iran [1]. Any infection that occurs within the first month of surgery at or near the operation site is defined as SSI. In the case of an implant operation, infections developed within the first year of implant insertion at the operation site are also known as SSI [2]. Despite major advances in surgical site sterilization and perioperative antibiotic prophylaxis, SSI is still a main complication in surgical practices [1]. Such infections can increase hospital length of stay, treatment cost, readmission, re-surgery, antibiotic consumption and even mortality in affected patients [3]. Moreover, it could affect the mental health of the patients and lead to anxiety, worrisome, and increased pain [4]. In most cases, the infection manifests as wound, cellulitis, and swelling, and it is not possible to prepare a specimen for culture. In other cases, due to the presence of pus and local abscess, the cause and sensitivity of the infection could be determined by the culture of the secretions.

Both Gram-positive and Gram-negative bacteria are known as the prevalent causes of SSI. Among the Gram-positive bacteria, Staphylococcus aureus, Enterococcus spp., and Streptococcus spp. and amongst Gram-negative organisms Pseudomonas aeruginosa, Escherichia coli (E. coli), Klebsiella spp., Proteus spp., Enterobacter spp. and Acinetobacter spp. were the main reported pathogens leading to SSI [5-17].

The global emergence of antibiotic resistance among bacteria has made the treatment of various infections challenging [18]. The prevalence of microbial resistance to antibacterials differs per hospital and is subject to constant change [20]. For instance, in a study that was conducted in Brazil in 2014-2017, Staphylococcus aureus, Enterobacter spp. and Pseudomonas aeruginosa were the predominant pathogens; all Gram-positive bacteria were sensitive to Vancomycin; and Gram-negative pathogens were more sensitive to Meropenem, Polymyxin, and Amikacin (6). While in a similar study in Pakistan that was carried out a year before the Brazilian survey, the most common pathogens were Staphylococcus aureus, E. coli, and Pseudomonas spp.; Linezolid and Vancomycin had the best antibacterial activity against Gram-positive bacteria; and the best antibiotics for treatment of Gram-negative organisms were Carbapenems, Piperacillin-Tazobactam and Cefoperazone [7].

In Iran, several studies have investigated the bacterial agents and the antibiotic susceptibility of SSI. In a study conducted in Yasuj in 2007, E. coli, Klebsiella spp., Pseudomonas spp. and Staphylococcus aureus were the most common etiologies and all the isolates showed high sensitivity to Ciprofloxacin and Gentamicin [8]. In another study conducted in Isfahan in 2008, Staphylococcus aureus, Klebsiella spp., E. coli, and Pseudomonas aeruginosa were the most common bacteria. The most effective antibiotics against Gram-negative and Gram-positive organisms were Imipenem/Meropenem, and Vancomycin respectively [9]. This study aims to update the information on microbial agents related to SSI and their antimicrobial susceptibility pattern in patients admitted to three major hospitals in Isfahan, Iran.

3. Materials and Methods

3.1. Study Design

This study was designed to report the major organisms that cause SSI and their antibiotic resistance pattern in the hospitalized patients in three referral hospitals in Isfahan, Iran, from 21st March 2016 to 20th March 2018. The hospitals that enrolled in the study were Al-Zahra, Dr. Shariati and Dr. Gharazi medical centers. The medical labs of these hospitals had a Quality Certificate for microbiological testing from the Iranian Ministry of Health and were a participant of the WHO (World Health Organization) in GLASS (Global Antimicrobial Resistance Surveillance System) program. This research is part of an extensive cross-sectional prospective surveillance conducted in those three mentioned hospitals, named as "Isfahan Antimicrobial Resistance Surveillance-1"(IAS-1) [20]. IAS-1 aimed to report the antibiotic susceptibility patterns of the causative agents of various infections in patients admitted in the three large medical centers mentioned above. The study also emphasized the inclusion of the true pathogens only and the exclusion of the contaminant ones [20]. The purpose of this study is to help health-care providers to choose the best antibiotics for the treatment of patients with SSI.

3.2. Specimen Collection

All the hospitalized patients with positive culture results from a surgical wound participated in this study. Surgical site infection was defined as the presence of soft tissue inflammation, edema, purulent discharge or fluid collection at or near a surgical incision site within the first month of surgery or within the first year of implant insertion. A single time growth of a common organism for SSI or the growth of an uncommon bacterium for SSI in two separate samples with identical susceptibility was considered as positive culture result. For sampling, the infected site was first scrubbed with normal saline and povidone-iodine and then culture specimen was obtained by swab or needle aspiration of material from the infected site. Specimens were transported to the laboratory within 20 minutes and generally inoculated within 1 hour after collection.

3.3. Bacterial Isolation and Antibiotic Susceptibility Testing

All collected specimens were processed to detect aerobe bacteria and antibiotic susceptibility patterns in accordance with the Clinical Laboratory and Standards Institute (CLSI) descriptions [21]. Dehydrated antibiotic discs from Mast, Merseyside, UK, and minimum inhibitory concentration (MIC) test strip from Liofilchem, Italy was used for antimicrobial sensitivity testing in all participating microbiology labs.

3.4. Data Collection and Analysis

Data on causative agents and their antibacterial sensitivity pattern, as well as gender and surgical service of participants were collected from WHONET v5.6 software. The result of antibacterial sensitivity was presented for all bacteria according to Gram staining category (Gram positive versus Gram negative) and for the most prevalent bacteria separately as raw numbers and percentages.

3.5. Ethical Consideration

The study was approved by the ethical committee of Isfahan University of Medical Sciences. (Approval number: IR.MUI.MED.REC.1399.341)

4. Results

Totally 90 patients with culture-positive SSI were detected, of which 54 (60%) were males, 51 (56.66%) admitted to the surgery ward, 13(14.4%) in orthopedic and 26(28.8%) in other wards.

Among the isolates, 60 were Gram-negative rods (66.6%) and 30 were Gram-positive cocci (33.3%). The most prevalent organism was E. coli (25.5%) followed by Staphylococcus aureus (22.2%), Klebsiella spp. (18.8%), Pseudomonas spp. (8.8%) and Enterococcus spp. (7.77%).

The antibiogram of the isolates showed that Gram-negative bacteria were highly sensitive to Imipenem and Aminoglycosides and highly resistant to Ciprofloxacin, Trimethoprim-Sulfamethoxazole and Third generation Cephalosporins. The antibiogram of Gram-positive organisms revealed high sensitivity to Rifampin and Vancomycin, moderate susceptibility to Aminoglycosides and Trimethoprim-Sulfamethoxazole; and low susceptibility to Erythromycin, Clindamycin, Tetracycline and Ciprofloxacin.

The Antibiotic sensitivity of the isolates in accordance to Gram staining as well as the species of the bacteria is presented in Table-1.

Table 1: The Antimicrobial sensitivity of isolates causing surgical site infection in the hospitalized patients in three referral hospitals in Isfahan, Iran in according to Gram staining and species of the bacteria

Antibacterial agent	Total Gram negatives	Total Gram positives	*E. coli*	*S. aureus*	*Klebsiella spp.*	*Acinetobacter baumanni*
Aminoglycosides	51/73(69.8%)	14/23(60.8%)	26/28(92.8%)	13/21(61.9%)	9/18(50%)	4/17(23.5%)
Tetracycline	-	7/20(35%)		5/15(33.3%)		-
Vancomycin	-	23/27(85.1%)		17/17(100%)		
Erythromycin	-	5/23(21.7%)		5/19(26.3%)		
Clindamycin	-	6/22(27.3%)		6/19(31.6%)		
Ciprofloxacin	14/56(25.0%)	10/26(38.5%)	7/22(31.8%)	9/19(47.4%)	1/16(6.2%)	0/10(0%)
Trimethoprim /Sulfamethoxazole	8/29(27.6%)	11/20(55%)	6/14(42.9%)	11/19(57.7%)	0/5(0%)	0/4(0%)
Rifampin	-	6/6(100%)		6/6(100%)		
Third generation Cephalosporins	21/57(36.8%)	-	13/26(50%)		1/17(5.9%)	0/12(0%)
Imipenem	13/17(76.5%)	-	9/9(100%)		1/3(66.7%)	1/6(16.7%)

5. Discussion

Our study revealed that SSI is more common in males than in females. In addition, we found that Gram-negative bacilli and Staphylococcus aureus are the main cause of this infection. Furthermore, the study showed that Gram-negative strains are more sensitive to Imipenem and Aminoglycosides and Gram-positive isolates are more susceptible to Vancomycin, Rifampin and Aminoglycosides.

In this study similar to earlier surveys, the prevalence of SSI in males was higher than in females [6, 9, 10, 14, 16]. This finding might be related to dominate occupation of males in activities such as agricultural, industrial or truck driving jobs that are more likely to be traumatic and require surgery.

In our study, E. coli was identified as the most common cause of SSI, followed by Staphylococcus aureus, Klebsiella spp., Acinetobacter spp., and Pseudomonas aeruginosa. In previous studies,

these bacteria have been among the most common causes of SSI [5-15]. However, in contrast to our results, Staphylococcus aureus was the primary cause of the infection in some former studies [6, 7, 9, 10-12, 15-17]. Furthermore, the prevalence of other Gram-negatives, including Klebsiella spp. [15], Acinetobacter spp. [16], Enterobacter spp. [6] and Pseudomonas aeruginosa [5, 10, 11] were higher than E. coli in some earlier researches. This discrepancy in various reports on the etiology of SSI might be related to the type and site of the surgery, the selected hospital's wards, the geographical location and the time frame of the research.

Moreover, in contrast to our findings, Coagulase-Negative Staphylococcus (CoNS) had been identified as a common cause of SSI in some previous investigations [5, 9, 11, 12, 15-17]. CoNS is part of the skin's normal inhabitant and can contaminate samples prepared from SSIs. In our investigation, the growth of CoNS in one surgical site sample was considered as a contamination and the specimen was excluded from the analysis. Therefore, in our study, this microorganism was not a main cause of SSI.

We found that isolated Gram-negative bacteria were highly susceptible to Imipenem and Aminoglycosides. In addition, among the isolates, E. coli and Klebsiella spp. had remarkable sensitivity to these class of antibiotics, whereas Acinetobacter spp. were highly resistant to all examined antibiotics, including above categories of antibiotics. In most similar studies, Gram-negative isolates had remarkable sensitivity to Carbapenems [5-7, 9, 14, 17]. However, in some earlier studies, the Gram negative isolates had reduced susceptibility to Carbapenems, albeit higher than other examined antibiotics [12]. This finding elucidates that this class of antibiotics could be effective in the treatment of SSI in most surgical centers. However, to prevent the emergence of resistant organisms, it is suggested to use this category of antibacterial only in the empiric treatment of severe and life-threatening SSIs; and to de-escalate therapy after determining the antibiotic sensitivity of the causative agent.

In contrast to our findings, in most of previous works, Gram-negative isolates had reduced susceptibility to Aminoglycosides [5, 10, 12, 16]. However, in some others, a high sensitivity of the isolates to this class of antibacterial was observed [6-8, 9, 11, 13, 14, 17]. This finding emphasized the significance of local, hospital-based surveillance of the bacterial spectrum and antibiotic sensitivity for logical empiric therapy of SSI.

In conformity with other reports, our results showed the high level of un-susceptibility of Gram-negative isolates to third-generation Cephalosporins [5-7, 9, 10, 12, 14, 16, 17] and Trimethoprim-sulfamethoxazole [8, 10, 11, 16]. This finding highlights that these antibiotics would not be suitable candidates for the empiric therapy of SSIs in most regions.

Our investigation in consistent with previous researches revealed that Gram-negative isolates had reduced sensitivity to Ciprofloxacin [5, 9, 10, 13, 14, 17]. However, in some other studies, high sensitivity of Gram-negatives to fluoroquinolones was reported [8, 16]. This inconsistency in susceptibility of Gram negatives among various reports is explained by the type and site of the surgery; as well as the locale and time frame of the research. This fact highlights the necessity of a surveillance system for antimicrobial resistance in each surgical center to determine the most effective antibiotics for the treatment of SSI in that hospital.

In this study, consistent with all previous studies, Staphylococcus aureus isolates was determined as the most prevalent Gram-positive bacterium that causes SSI [5-17]. Moreover, our findings as well as many of similar researches revealed that all isolated Staphylococcus aureus were sensitive to Vancomycin [5, 6, 16, 17]. However, in some studies [7, 9, 12, 14] remarkable resistance of Staphylococcus aureus isolates to vancomycin had been reported. Due to the rarity of resistance to Vancomycin among Staphylococcus aureus isolates [22]; it seems that this significant prevalence of resistance in some earlier researches could be related to technical errors in those studies. The remarkable susceptibility of Staphylococcus aureus strains to Vancomycin indicates that this antibiotic would be a suitable choice for inclusion in the treatment of SSI in studied hospitals.

Our study revealed that all Staphylococcus aureus isolates were susceptible to Rifampin. This finding indicates that this drug can be used as an adjunctive to Vancomycin in severe and life-threatening SSIs in above hospitals. In contrast to our results, in most previous studies that have investigated the sensitivity of Staphylococcus aureus to this drug, remarkable un-susceptibility to this antibiotic had been observed [5, 7, 9, 15].

Consistent with previous studies, we found a high level of un-susceptibility of Gram-positive strains to Trimethoprim-sulfamethoxazole [10, 14], Clindamycin [5, 7, 10, 14, 16] Erythromycin [5, 7, 10, 11, 14] and Tetracycline [5, 9, 11, 14]. This finding indicates that these antibiotics are no longer suggested for SSI treatment in most geographic areas.

Our study had two major limitations. First of all, the number of enrolled patients in the study was limited. As previously noted in most investigations on SSI there is no purulent specimen for culturing. Consequently, in many of the similar studies, the number of specimens was limited [10-15]. Second of all, this research was conducted in limited medical centers and generalization to other surgical centers could not be done.

In conclusion, our study revealed that Gram-negative bacilli and Staphylococcus aureus are the most common causes of SSI in the studied hospitals. Moreover, the sensitivity of Gram-negatives to Imipenem and Aminoglycosides and the sensitivity of Gram-positives to Vancomycin and Rifampin were remarkably high. Therefore, these drugs might be the most appropriate antibiotics in treating such infections. High resistance of Gram-negatives to

third-generation Cephalosporins and Ciprofloxacin; and high level of resistance among Gram-positives to Clindamycin, Erythromycin, and Tetracycline indicate that these antibiotics would have little effect in the treatment of such infections. Surveillance of antimicrobial resistance and rational use of antibiotics is suggested for the effective treatment of the infection as well as the prevention of microbial resistance in all hospitals.

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