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EVALUATION OF POSTOPERATIVE WOUND CULTURE RESULTS AT AN ORTHOPEDIC CLINIC: A THREE-YEAR ANALYSIS

ORTOPEDİ KLİNİĞİNDE CERRAHİ OPERASYON SONRASI YARA KÜLTÜR SONUÇLARININ DEĞERLENDİRİLMESİ: ÜÇ YILLIK ANALİZ

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Abstract

Objective: Wound infections constitute the most prevalent postoperative complication encountered by orthopedists. Lack of timely and correct treatment may lead to chronic wound infections. Therefore, this study aims to guide empirical treatment, investigate the isolated bacteria and associated antibiotic resistance states in wound cultures obtained after orthopedic surgery.
Methods: In the three-year period between January 2016 and end of December 2018, 3100 patients were operated in the Orthopedics and Traumatology Clinic of the Kirsehir Ahi Evran University Training and Research Hospital. Wound cultures obtained from patients operated due to wound infections were retrospectively evaluated. Antibiotic susceptibility tests were performed in accordance with EUCAST guidelines using disc diffusion methods and the VITEK 2 Compact® (Biomerieux, France) automated system.
Results: A total of 284 wound culture specimens were evaluated, and 85 (22.9%) of these demonstrated significant bacterial growth. Monomicrobial growth was detected in 92% and polymicrobial growth was found in 8% of the cultures. The most commonly isolated microorganism was *Escherichia coli* (22.3%), followed by coagulase negative staphylococci (CoNS) (20%), *Staphylococcus aureus* (15.3%), *Acinetobacter baumannii* (9.4%), *Pseudomonas aeruginosa* (9.4%) and *Streptococcus spp.* (5.8%) respectively. Extended-spectrum beta-lactamase (ESBL) positivity was determined as 100% and 25% for *E. coli* and *K. pneumoniae* isolates, respectively. Carbapenems were found to be the most effective antibiotics to *Enterobacteriales* family. Gram positive bacteria were not resistant against vancomycin and teicoplanin, but 7.6% of *S. aureus* strains and 57.1% of CoNS were methicillin-resistant.
Conclusion: Identification and monitoring of causative microorganisms in postoperative wound infections and antibiotic resistance rates at every hospital is quite important. Taking these findings into consideration will result in better prevention of infections, increased treatment success, and decreased antibiotic resistance rates.

Keywords: *Orthopedics, infection, wound, culture, antibiotic resistance*

Öz

Amaç: Yara enfeksiyonları ortopedistlerin cerrahi sonrası karşılaştıkları komplikasyonların başında gelmektedir. Hızlı ve doğru tedavi uygulanmaz ise kronik yara enfeksiyonları gelişebilmektedir. Bu nedenle ampirik tedaviye yol göstermesi amacıyla, çalışmamızda ortopedik cerrahi sonrası yara kültürlerinden izole edilen bakterilerin dağılımı ve antibiyotik direnç durumları retrospektif olarak değerlendirilmiştir.
Yöntem: Ocak 2016- Aralık 2018 sonu arasındaki üç yıllık dönemde Kırşehir Ahi Evran Üniversitesi Eğitim ve Araştırma Hastanesi ortopedi ve travmatoloji kliniğinde 3100 hasta opere edilmiştir. Opere edilen hastaların yara enfeksiyonu nedeni ile alınan yara kültürleri retrospektif olarak incelenmiştir. İzole edilen mikroorganizmaların antibiyotik duyarlılık testleri EUCAST önerileri doğrultusunda disk diffüzyon yöntemi ve VİTEK 2 Compact® (Biomerieux, France) otomatize sistem ile çalışılmıştır.
Bulgular: Değerlendirilen 284 yara kültürü örneğinin 85'inde (%29,9) anlamlı üreme olmuştur. Kültürlerin %92'sinde monomikrobiyal, %8'inde polimikrobiyal üreme saptanmıştır. En sık izole edilen mikroorganizma *Escherichia coli* (%22,3) olup ve bunu sırasıyla Koagülaz Negatif Stafilokoklar (KNS) (%20), *Staphylococcus aureus* (%15,3), *Acinetobacter baumannii* (%9,4), *Pseudomonas aeruginosa* (%9,4), *Streptococcus spp.*'nin (%5,8) takip ettiği belirlenmiştir. Genişlemiş spektrumlu betalaktamaz (GSBL) pozitifliği *E. coli* ve *K. pneumoniae* izolatları için sırasıyla %100 ve %25 oranında belirlenmiştir. *Enterobacteriales* ailesine en etkili antibiyotik grubunun karbapenemlerin olduğu görülmüştür. Gram pozitif bakterilerde vankomisin ve teikoplanin direnci bulunmamakla birlikte metisilin direnci *S. aureus* suşlarının %7,6'sı ve KNS'ların %57,1'inde tespit edilmiştir.
Sonuç: Her hastanede, cerrahi sonrası gelişen yara enfeksiyonlarında etken mikroorganizmaların ve antibiyotik direnç oranlarının bilinmesi gerekir. Bu verilerin dikkate alınması ile enfeksiyonların önlenmesi, tedavi başarılarının artması ve antibiyotik direnç gelişiminde azalma olabileceğini düşünmekteyiz.

Anahtar Kelimeler: *Ortopedi, enfeksiyon, yara, kültür, antibiyotik direnci*

Introduction

Wound infections develop due to the impairment of skin integrity following trauma and the invasion of the wound site by pathogenic microorganisms.[1] Infections of the wound site result in chronic wounds and delay recovery.[2] There has been a marked increase in the number of orthopedic surgery cases. In spite of improved sterilization conditions and antisepsis practices, and the use of prophylactic antibiotics, the postoperative complication that causes the greatest concern for orthopedic surgeons remains to be wound infections.[3] Orthopedic surgical site infection is defined as the microbial contamination of the surgical site within 30 postoperative days, or one year if an implant was placed.

Infections encountered during the postoperative wound recovery period may originate from the patient's own flora or have a nosocomial origin. Nosocomial causative microorganisms are more resistant against antibiotics.[4] The difficulty of the treatment of resistant microorganisms results in increased morbidity and mortality, prolonged hospitalization, and a higher treatment cost. Knowledge on the distribution of common causative agents of wound infections and associated antibiotic susceptibilities are important as it would guide empirical treatment and prevent antibiotic resistance by promoting correct use of antibiotics based on resistance data.[5]

This study aims to investigate the microorganisms isolated from wound infections, associated antimicrobial resistance rates and to guide empirical treatment in patients postoperatively hospitalized in the orthopedics clinic and outpatients who presented to the polyclinic at a later postoperative stage.

Methods

Microorganisms isolated from 284 wound culture specimens that were sent to the Kırşehir Ahi Evran University Training and Research Hospital Medical Microbiology laboratory from the Orthopedics and Traumatology Clinic between January 2016-December 2018 and associated antibiotic resistance rates were retrospectively evaluated.

Swab, soft tissue, and aspirated fluid samples obtained from wound sites were assessed for leukocyte and microorganism presence by preparing a Gram stain preparation. Samples were simultaneously inoculated onto 5% sheep blood agar, chocolate agar, and eosin methylene blue agar (EMB) media (RDS med, Turkey), and were incubated for 48 hours at 37°C. A VITEK 2 Compact® (bioMérieux, France) fully-automated identification system was used for the identification of bacterial growth and antibiotic susceptibility. Additionally, in situations where the automated system could not be utilized, the catalase, coagulase, and PYR tests were used for gram positive bacteria, and conventional methods such as motility, indole, urea, and oxidase were used for gram negative bacteria. Antibiotic susceptibility tests were performed with the Kirby-Bauer disc-diffusion method by spreading bacterial suspension adjusted to 0.5 McFarland over the surface of a Mueller Hinton agar medium. The presence of ESBL in *E. coli* and *K. pneumoniae* isolates was investigated by double disc synergy method. Tests were performed and evaluated in accordance with EUCAST recommendations.[6] *E. coli* ATCC 25922 and *S. aureus* ATCC 29213 were used as a reference strain in antimicrobial susceptibility testing.

Coagulase negative staphylococci (CoNS) have been reported in invasive specimens if polymorphonuclear leukocytes were abundant in gram staining. If cultures showed mixed bacterial growth with significant presence of epithelial cells, specimens were evaluated and reported as elements of the normal skin flora.

Results

Of the total 284 wound site specimens collected over the three-year period under investigation, 248 (87.3%) were obtained from patients hospitalized in the orthopedics and traumatology service, while 36 (12.7%) were obtained from patients who presented to the polyclinic. The number of wound culture specimens demonstrated significant bacterial growth was 85 (29.9%) and eight (2.8%) contamination with normal skin flora. Of the cultures, 92% were determined to be monomicrobial growth and %8 to be polymicrobial growth. Of the 85 isolated agents, 47 (55%) were determined to be gram negative bacteria and 38 (45%) to be gram positive bacteria. Of the agents, 11 (13%) were isolated from specimens collected from outpatients and 74 (87%) from specimens of hospitalized patients. *E. coli* (n=19, 22.3%) and CoNS (n=17, 20%) were the most commonly isolated bacteria, followed by *Staphylococcus aureus* (n=13, 15.3%), *Acinetobacter baumannii* (n=8, 9.4%), *Pseudomonas aeruginosa* (n=8, 9.4%) and *Enterobacter cloacae* (n=5, 5.8%) respectively. The distribution of CoNS assessed and reported as agents can be listed in descending order of prevalence as; *S. epidermidis* (n=8, 47%), *S. haemolyticus* (n=5, 29.5%), and *S. lugdunensis* (n=4, 23.5%). The distribution of bacteria that demonstrated growth has been presented in Table 1. Methicillin resistance was determined in one (7.6%) of the isolated *S. aureus* strains and ten (57.1%) of isolated CoNS. All *E. coli* isolates were ESBL positive, however, carbapenem resistance was not detected. In *K. pneumoniae* isolates, ESBL positive was detected in one (25%) isolate. All of the *A. baumannii* isolates were resistant against ciprofloxacin and carbapenems. Rates of resistance were 75% against aminoglycosides and 37.5% against trimethoprim sulfamethoxazole. Meanwhile, all isolates were susceptible to the antibiotics tigecycline. Resistance rates against various antibiotics have been presented in Table 2 for gram positive bacteria, and in Tables 3 and 4 for gram negative bacteria.

Table 1. Distribution of microorganisms identified in post orthopedic surgery wound cultures

Microorganisms	**n**	**%**
E. coli	19	22.3
Coagulase negative *Staphylococci*	17	20
S. aureus	13	15.3
A. baumannii	8	9.4
P. aeruginosa	8	9.4
Streptococcus spp.	5	5.8
E. cloacae	5	5.8
K. pneumoniae	4	4.7
Enterococcus spp.	3	3.6
Serratia marcescens	2	2.4
Morganella morganii	1	1.2
Total	85	100

Discussion

Postoperative wound infections constitute one of the most important problems in orthopedic surgery.[7] Surgical site infections are caused by microorganisms in the normal flora that are found in or carried to the surgical site, and thus, most of these infections have an endogenous origin.[8] Although components of the endogenous flora are usually non-pathogenic, they can become pathogenic if they are allowed to contaminate the wound. Risk factors associated with orthopedic infections include various factors such as the clinical condition of the patient, a long preoperative hospitalization time, operative time, skin preparation, hand sanitization technique of the surgeon and the surgical team, conditions of the operating room environment, number of people in the operating room, the technique of the surgeon, and use of implants.[9-12] Surgical wound infections are among the leading causes of postoperative mortality and morbidity, and delayed recovery prolongs hospitalization and increases treatment cost.[3]

Table 2. The number and the rates of antimicrobial resistance in gram positive bacteria

Antibiotics	*S. aureus* n (%)	*CoNS* n (%)	*Enterococcus spp.* n (%)
Penicillin	11 (84.6)	17 (100)	NT
Ampicillin	NT	NT	2 (66.6)
Methicillin	1 (7.6)	10 (57.1)	NT
Clindamycin	0	7 (41)	NT
Daptomycin	0	0	0
Erythromycin	1 (7.6)	7 (41)	NT
Fusidic acid	0	0	NT
Gentamicin	0	4 (23.5)	1 (33.3)
Ciprofloxacin	0	6 (35)	0
Levofloxacin	0	0	NT
Linezolid	0	0	0
Mupirocin	0	0	NT
Tetracycline	2 (15.3)	6 (35.2)	NT
TMP-SMX	0	3 (17.6)	0
Teicoplanin	0	0	0
Vancomycin	0	0	0

TMP-SMX, Trimethoprim-sulfamethoxazole*;* NT, Not tested.

Table 3. The number and the rates of antimicrobial resistance in *Enterobacteriales*

Antibiotics	*E. coli* n (%)	*K. pneumoniae* n (%)	*E. cloacae* n (%)
Ampicillin	19 (100)	4 (100)	5 (100)
AMC	11 (58)	2 (50)	5 (100)
Cefuroxime	19 (100)	1 (25)	5 (100)
Ceftriaxone	19 (100)	1 (25)	0
Cefotaxime	19 (100)	1 (25)	0
Cefepime	16 (84.2)	1 (25)	0
Gentamicin	3 (15.8)	1 (25)	0
Amikacin	0	0	0
Ciprofloxacin	19 (100)	1 (25)	0
Levofloxacin	5 (26.3)	1 (25)	0
Imipenem	0	0	0
Meropenem	0	0	0
Ertapenem	0	0	0
PTZ	4 (21)	3 (75)	0
TMP-SMX	19 (100)	1 (25)	0

AMC, amoxicillin clavulanate; PTZ, Piperacillin-tazobactam; TMP-SMX, Trimethoprim-sulfamethoxazole; NT, Not tested.

There are several studies in the literature that have investigated postsurgical wound culture results.[3,7,12] A study published by Savci et al.[7] in 2018 determined the type of bacteria isolated most commonly from wound cultures obtained after orthopedic surgery as *S. aureus* 38.5%, followed respectively by *E. coli* %26.3, *E. cloacae* 12.3%, *A. baumannii* 5.3%, *P. aeruginosa* 5.3%, *K. pneumonia*e 3.5%, *S. haemolyticus* 3.5%, *E. aerogenes* 1.8%, *E. faecalis* 1.8%, and *E. americana* 1.8%. They reported that the most microorganisms were isolated after hip surgery. In another study, microorganism was isolated in 37% of the wound specimens of patients suspected to have infection after orthopedic surgery. Isolated bacteria were listed in descending order as *Acinetobacter spp.* 25%, *P. aeruginosa* 20%, *S. aureus* 16%, *E. coli* 10%, *K. pneumoniae* 10%, CoNS 8%, *P. mirabilis* 5%, *Enterobacter spp.* 4%, and *Enterococcus spp.* 1%.[3] In a study Doshi et al. [12] published in 2017, bacterial growth in wound infections encountered after tibia fracture surgery included *S. aureus* at a rate of 81%, *P. aeruginosa* at 9,1%, and *Enterobacter spp* growth at 9,1%. Similarly, Cirit et al. [5] reported that CoNS were the most common bacteria isolated from wound cultures and this was followed by *E. coli*, *S. aureus*, *P. aeruginosa*, *A. baumannii*, and other enteric bacteria. A 2014 study conducted in Van investigated wound cultures collected over a three-year period and presented the distribution of bacterial growth in descending order as *A. baumannii* 24%, *P. aeruginosa* 12%, *S. aureus* 11%, *E. coli* 10%.[13] In our study, *E. coli* and CoNS were the most commonly isolated bacteria and they were followed by *S. aureus*, *A. baumannii*, *P. aeruginosa*, and *E. cloacae*. When compared, our results appear to be in accordance with the results of other studies.

Table 4. The number and the rates of antimicrobial resistance in non-fermentative gram-negative bacteria

Antibiotics	*A. baumannii* n (%)	*P. aeruginosa* n (%)
Ceftazidime	NT	1 (12.5)
Cefepime	NT	0
Gentamicin	6 (75)	0
Amikacin	5 (62.5)	1 (12.5)
Netilmicin	6 (75)	1 (12.5)
Ciprofloxacin	8 (100)	0
Levofloxacin	8 (100)	0
Imipenem	8 (100)	0
Meropenem	8 (100)	0
PTZ	NT	4 (50)
TMP-SMX	3 (37.5)	1 (25)
Colistin	0	0
Tigecycline	0	NT

PTZ, Piperacillin tazobactam; TMP-SMX, Trimethoprim-sulfamethoxazole; NT, Not tested.

It is very important to perform the infection prophylaxis consciously and effectively. Incomplete or incorrect treatment may result in bacterial resistance.[10] Increases in bacterial antibiotic resistance are concerning as they may result in treatment failure, prolonged hospitalization, and nosocomial infections.[14] A review of the literature with respect to antibiotic resistance rates of bacteria that grow in postoperative wound infections reveals investigations of methicillin resistance in gram positive bacteria. Savci et al. [7] determined methicillin resistance rates of *S. aureus* and CoNS isolated from wound cultures following orthopedic surgery as 9% and 100%, respectively. In another study that investigated post-orthopedic surgery wound infections, Gormeli et al. determined these resistance rates as 7% and 50%, respectively.[3] In other studies, methicillin resistance

rates for *S. aureus* and CoNS were reported respectively as 29% and 50% by Yurtsever et al.[15], 20% and 8% by Polat et al.[16], 22% and 33% by Gundem et al.[2] In our study, 7.6% of isolated *S. aureus* strains and 57.1% of CoNS were found to be methicillin resistant. We find that our methicillin resistance is consistent with the results of other studies.

In the recent years, antibiotic resistance has increased significantly among gram negative microorganisms that cause nosocomial infections.[17] Particularly, carbapenem resistance in the *Enterobacteriales* group and colistin resistance in *A. baumannii* reduce treatment options significantly.[17,18] However, carbapenem resistance did not determine in our study in the *Enterobacteriales* group of gram negative bacteria that manifested growth after orthopedic surgery, although the same group was positive for ESBL (*E. coli* 100%, *K. pneumoniae* 25%). Our *P. aeruginosa* isolates were highly susceptible to antibiotics and did not have high resistance rates. Among other studies, Gormeli et al.[3] did not determine carbapenem resistance in the *Enterobacteriales* group, meanwhile, reported 92.2% carbapenem resistance in *A. baumannii*. All strains showed colistin susceptibility. ESBL rates for *E. coli* and *K. pneumoniae* were respectively reported as 10% and 40%. Savci et al.[7] determined carbapenem resistance rates of 26.7% for *E. coli* and 25% for the *Enterobacter spp*. group. While they did not provide ESBL rates, they determined cefepime resistance rates as 93.5% for *E. coli* and 37.5% for *Enterobacter spp*. As stated in the same study, resistance rates of *A. baumannii* isolates showed a rapid increase over the years, resulting carbapenem resistance rates as high as 92%.[7]

Managing surgical infections and taking preventative measures require the identification of infection risk factors associated with the host, microorganism, environment, and implant material.[19]

Limitation

The limitations of our study include its retrospective nature and our inability to discriminate between skin and soft tissue infections. Our opinion is that solution oriented prospective studies are warranted that consider the types of surgical operation, operative durations, elective and urgent surgeries, assess the knowledge of nurses and other health practitioners on surgical site infections, investigate extensive risk factors, and evaluate the parameters concerning the quality of patient care.

Conclusion

The majority of wound specimens that are referred to the microbiology laboratory in particular are constituted by wound culture specimens obtained after orthopedic surgery.[20,21] Unfortunately, training of health personnel and compliance in practice are not yet ideal in developing countries. In order to prevent contamination with the skin flora, optimal preoperative asepsis and antisepsis conditions must be met and postoperative infections must be prevented effectively by providing postoperative care as meticulously. Particularly following surgical interventions close to the perianal region, sanitization and antisepsis guidelines must be followed with enough care in order to prevent infections originating from intestinal flora. In addition, the necessity of keeping hygiene guidelines, especially hand sanitization, must be stressed to the patients and hospital personnel through continuous training. In order to achieve these standards, constant monitorization of infection rates, agents, and antibiotic resistance rates at our orthopedics clinic will serve as a guidepost.

Conflict of Interest

The authors have no conflicts of interest to declare.

Compliance with Ethical Statement

The research was approved by the Kırşehir Ahi Evran University Ethical Committee (2019-16/169).

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Author Contributions

HS, FMS: Design; HS: Project development; HS, FMS: Data collection; HS, FMS: Analysis; HS, FMS: Literature search; HS: Manuscript writing

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