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Postoperative Nosocomial Infections and Antimicrobial Resistance Pattern of Bacteria Isolates among Patients Admitted Al Jalla Hospital

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ABSTRACT

Hospital-acquired infections constitute a major public health problem worldwide. Postoperative wound infection can occur on the first day but commonly occurs between the fifth and tenth days after surgery. This study aimed to determine the prevalence of nosocomial pathogens among patients with postoperative wound infections at Al Jalla Hospital in Benghazi, along with the antibiotic susceptibility pattern of pathogens. Wound swabs were collected from 49 patients from May 1, 2023, to May 31, 2023. The cultivation of the sample used three types of media: MacConkey agar, blood agar, and chocolate agar. The inoculated agar plates were incubated aerobically at 370 °C overnight. The other patient's swab was used for Gram staining smears to make a presumptive diagnosis based on microscopic inspection. Identification of cultured isolates was done according to standard bacteriological techniques. A total of 100 bacterial isolates were identified from surgical sites. Of these isolates, Klebsilla pneumonia and Acinetobacter baumanni were the most frequently detected bacteria (19%), followed by Pseudomonas aeruginosa (13%), E. coli (7%), Enterobacter cloacae (6%), Proteus mirabbilis, Bacillus spp., and Pseudomonas putida (5%), Staphylococcus albus (4%), Staphylococcus aureus, Streptococcus pyogenes, and Chryseomonas luteola (3%), while Ochrobacter antihropi, Enterobacter sakazakii, Pantoea agglomerans, Sarratia marcescens, Klebsilla oxytoca, and Citrobacter spp. In conclusion, Al Jalla Hospital needs to make a concerted effort to minimise those infections by following strict aseptic operation procedures, effective methods of sterilisation, and patient management, as the bacterial isolates detected in our patients were terribly resistant to commonly available and prescribed antimicrobial drugs.

1. INTRODUCTION

A nosocomial infection can be defined as an infection acquired in a hospital by a patient who was admitted for a reason other than that infection. An infection occurs in a patient in a hospital or other healthcare facility in whom the infection was not present or incubating at the time of admission, and this includes infections that are acquired in the hospital but appear after discharge (WHO, 2010). Hospital-acquired infections (HAIs) constitute a major public health problem worldwide. They result in major causes of morbidity and mortality, functional disability, emotional suffering, and economic burden among hospitalised patients (Endalafer et al., 2011; Kamat et al., 2008). The most common types of nosocomial infections that could occur in a hospital setting are surgical wound and other soft tissue infections, urinary tract, respiratory, and blood stream infections (Graves, 2000).

Postoperative wound infection can occur from the first day onwards to many years after an operation, but commonly occurs between the fifth and tenth days after surgery (Medical Disability Guidelines, 2010). It may originate during the operation, i.e., as a primary wound infection, or it may occur after the operation from sources in the ward or as a result of some complications, i.e., a secondary wound infection (Pradhan & Agrawal, 2009). Most postoperative wound infections are hospital-acquired and vary from one hospital to the next and even within a given hospital, and they are associated with increased morbidity and mortality (Isibor et al., 2008). The site of infection may be limited to the suture line or may become extensive in the operative site, and the infecting microorganisms are variable, depending on the type and location of surgery and the antimicrobials received by the patient (WHO, 2010). Surgical site infections (SSIs), which account for 17% of all healthcare-associated infections, are the second most common HAIs next to urinary tract infections. They occur after approximately 3% of all operations and result in greater lengths of stay and additional costs (Napolitano, 2010). On the other hand, nosocomial bloodstream infections (BSIs), which represent 14% of total nosocomial infections, can be classified as primary or secondary. Primary nosocomial bacteremia occurs without any infection in or on other sites, whereas secondary bacteremia is the presence of infection in sites such as the urinary tract, a surgical wound, or the lower respiratory tract, which can lead to a bloodstream infection with the same organism (Pradhan & Agrawal, 2009). The emergence of polyantimicrobial-resistant strains of hospital pathogens has also presented a challenge in the provision of good quality in-patient care (Kamat et al., 2008). The battle between bacteria and their susceptibility to drugs is still problematic among the public, researchers, clinicians, and drug companies that are looking for effective drugs (Biadglegne et al., 2009). Postoperative wound infection by resistant bacteria worsens the condition, and it has become a serious problem in developing countries owing to poor infection prevention programmes, crowded hospital environments, and irrational prescription of antimicrobial agents. So this study was to isolate bacterial pathogens from hospital-acquired surgical sites and bloodstream infections and determine their current antimicrobial resistance patterns among patients.

2. METHOD

Patients

Swabs were collected from 49 patients in at Al Jalla Hospital in Benghazi City. A hospital-based postoperative study was conducted from May 1, 2023, to May 31, 2023The swabs were collected from the Intensive Care Unit in 8 cases, the Thoracic Surgery Ward in 5 cases, the Outpatients Department in 6 cases, the Male Surgery Ward in 4 cases, the Neurosurgery Ward in 4 cases, the Female Orthopaedics Ward in 2 cases, the Female Surgery Ward in 8 cases, and the Male Orthopaedics Ward in 3 cases.**Cultivation of sample**

Used three types of media, such as MacConkey agar, blood agar, and chocolate agar, and these media are best prepared from ready-to-use dehydrated powder, available from most suppliers of culture media.

Identification of Isolated bacteria

- **Gram Stain**

The Gram staining reaction is used to help identify pathogens in specimens and cultures by their Gram reaction (Gram positive or Gram negative) and morphology.

- **Biochemical tests**

Catalase test

This test is used to differentiate those bacteria that produce the enzyme catalase, such as staphylococci, from non-catalase producing bacteria such as streptococci.

Coagulase test

The coagulase test is used to differentiate Staphylococcus aureus (positive) from coagulase-negative staphylococcus (CONS). Coagulase is an enzyme produced by S. aureus that converts (soluble) fibrinogen in plasma to (insoluble) fibrin. Two types of coagulases are produced by most strains of Staphylococcus aureus

DNAase test

This test is used to help in the identification of S. aureus which produces deoxyribonuclease (DNAase) enzymes. The DNAase test is particularly useful when plasma is not available to perform a coagulase test or when the results of a coagulase test are difficult to interpret

Citrate utilization test

This test is one of several techniques used occasionally to assist in the identification of enterobacteria. The test is based on the ability of an organism to use citrate as its only source of carbon.

Indole test

Testing for indole production is important in the identification of enterobacteria. Most strains of E. coli, P. vulgaris, P. rettgeri, M. morganii, and Providencia species break down the amino acid tryptophan with the release of indole.

Oxidase test

The oxidase test is used to assist in the identification of Pseudomonas, Neisseria, Vibrio, Brucella, and Pasteurella species, all of which produce the enzyme cytochrome oxidase.

Urease test

Testing for urease enzyme activity is important in differentiating enterobacteria. Proteus strains are strong urease producers. Y. enterocolitica also shows urease activity (weakly at 35–37 ^{0}C). Salmonellae and shigella do not produce urease.

Triple sugar iron agar (TSI)

Triple sugar iron is a differential medium that contains. Three sugars Lactose1%, sucrose 1%, a small amount of Glucose 0.1%. Ferrous sulfate to detect H2s formation. Phenol red: the pH indicator. It is used to differentiate enteric based on the ability to reduce sulfur and ferment carbohydrates (Identification of G- ve bacteria). The medium is inoculated on the slant surface and also stabbed right to the butt of the tube and incubated for 24 h.

- **Analytical Profile Index (API 20E)Test**

This test was used for identification of Enterobactraiceae

- API 20E presented herein is a biochemical panel for identification and differentiation of members of the family Enterobacteriaceae. See fig. 1.

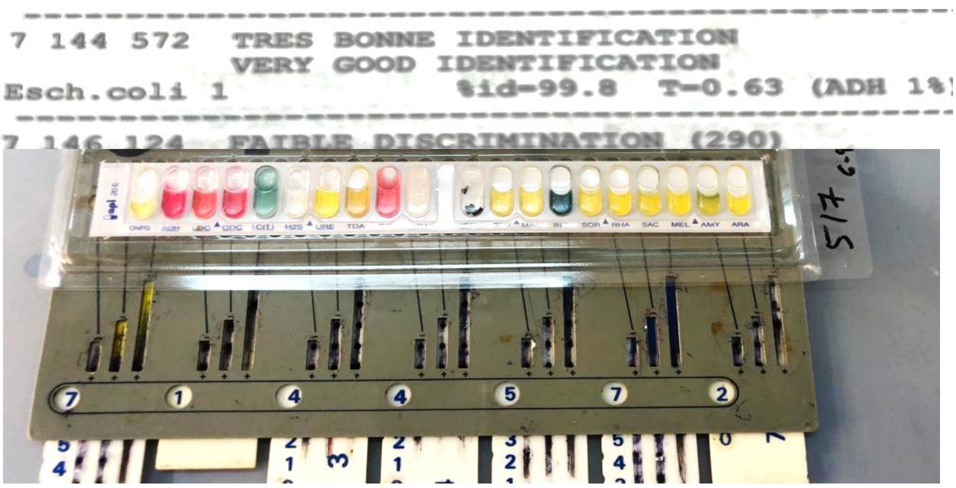

Figure 1. Analytical Profile Index (API 20E) Test

For investigation, data will be collected Two wound specimens were collected aseptically using sterile cotton swabs by experienced laboratory personnel, and the swabs were immediately dipped into a sterile tube containing two drops of sterile normal saline and delivered to Al Jalla Hospital Laboratory within five minutes of collection. Then, one of the wound swabs was inoculated on blood agar, MacConkey agar, and chocolate agar (all Oxoid, Ltd., England). The inoculated agar plates were incubated aerobically at 37 °C overnight. The other wound swab was used for Gram staining smears to make a presumptive diagnosis and to select a significant organism based on the quantitative measurements made on microscopy. Identification of cultured isolates was done according to standard bacteriological techniques. Antimicrobial susceptibility testing was performed using the Kirby Bauer agar disc diffusion technique for the isolated pathogen. A loop full of bacteria was taken from a pure culture colony and transferred to a tube containing 5 ml of phosphate buffered saline and mixed gently until it formed a homogenous suspension, and the turbidity of the suspension was adjusted to the turbidity of the McFarland 0.5 standard in a tube. The standardised inoculums of each isolate were inoculated on Mueller-Hinton antibiotic sensitivity medium (Oxoid, Ltd., England). Finally, all the isolates were tested for these underlisted Oxoid drug discs: Ampicillin (AP, 10μg), Penicillin G (P, 10IU), Amoxicillin (AML, 20μg), Chloramphenicol (C, 30μg), Gentamycin (CN, 10μg), Tetracycline (T, 30μg), Trimethoprim-sulphamethoxazole (Ts, 25μg), Ceftriaxone (CRO, 30μg), Doxycycline (D, 30μg), Norfloxacin (NOR, 10μg), Ciprofloxacin (CIP, 5μg), Nalidixic acid (NA, 30μg), Erythromycin (E,15μg), Kanamycin (K,30μg) and Nitrofurantoin (F, 300μg). These antimicrobial drug discs were selected based on the Clinical and Laboratory Standards Institute (CLSI), the availability, and prescription frequency of these drugs in the study area. The plates were incubated aerobically at 37 0C for 18-24 hours and the interpretation of the results of the antimicrobial susceptibility was made based on the CLSI criteria as sensitive, intermediate and resistant by measuring diameter of inhibition the zone (19).

All intermediate readings were taken as resistant during data entry. The standard reference strains, Staphylococcus aureus (ATCC25923), Escherichia coli (ATCC25922) and P. aeruginosa (ATCC 27853) were used to assure testing performance of the potency of drug discs as well as quality of culture media. The quantitative data was checked for completeness, coded and fed into excel.

3. RESULT

A total of 49 patients were undergone major operations and admitted in Intensive Care Unit (n=18), Thoracic surgery ward (n=7), Out patients Department (n=7), Males surgery ward (n=3), Neurosurgery ward (n=3), Females Orthopedics ward (n=1), females surgery ward (n=6) and Males Orthopedics ward (n=4). The sex of these patients showed that 36 (73.4%) were males and 13(26.5%) were females the mean age of patients was 32.2 years and 43 (87.7%) of them were older than 15 years. A total of 100 bacterial isolates were identified from surgical sites. Of these isolates, Klebsilla pneumonia and Acinetobacter baumanni were the most frequently detected bacterium (19%) followed by Pseudomonas aeruginosa was (13%), E.coli was (7%),Enterobacter cloacae (6%), Proteus mirabbilis and Bacillus spp. And Pseudomonas putida Were (5%), Staphylococcus Albus was (4%), Staphylococcus aureus , Streptococcus pyogenes and chryseomonas luteola were (3%), while Ochrobacter antihropi, Enterobacter sakazakii, Pantoea agglomerans, Sarratia marcescens, Klebsilla oxytoca and Citrobacter spp. were (1%). As shown in fig. 2.

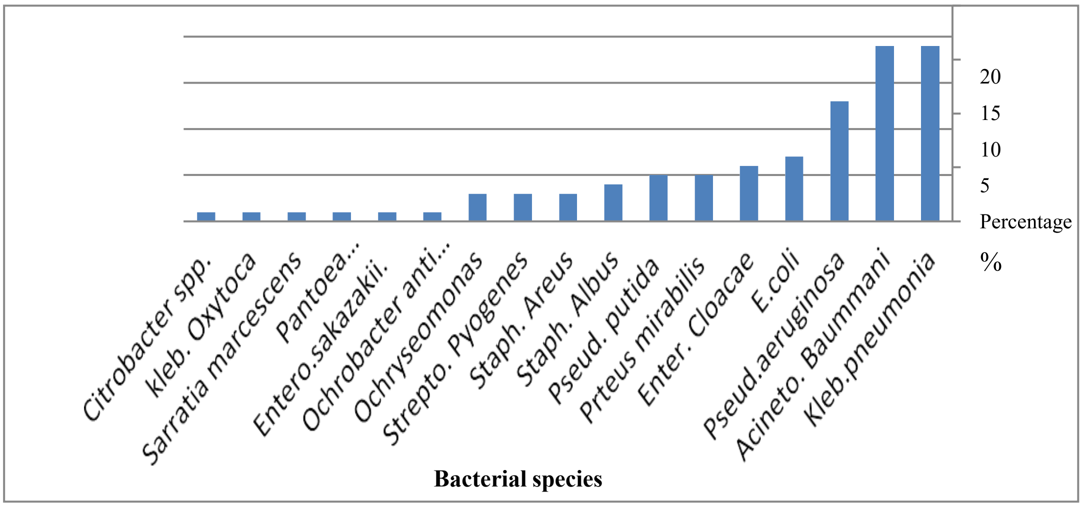

Figure 2. Frequency of nosocomial pathogenic bacterial isolates from postoperative patients

Pathogens has been isolated 35 from 18 specimens from Intensive Care Unit, The most common pathogens as follows; Acinetobacter baumanni was 15 (42.86%) , Pseudomonas aeruginosa was 6 (17.14%), Klebsilla pneumonia was 5(14.29%), E.coli was 3(8.57%), Proteus mirabbilis was 2(5.71%), Bacillus spp. was 2(5.71%), , Sarratia marcescens and chryseomonas luteola were 1 (2.86%). as shown in table 1.

Table 1. Common pathogens isolated from Intensive Care Unit

Pathogen	Number	Percentage %
Acinetobacter baumanni	15	42.86
Pseudomonas aeruginosa	6	17.14
Klebsilla pneumonia	5	14.29
E.coli	3	8.57
Proteus mirabbilis	2	5.71
Bacillus spp.	2	5.71
Sarratia marcescens	1	2.86
chryseomonas luteola.	1	2.86
Total	Total = 35	100

The antimicrobial drug resistance profile of bacterial isolates showed that Pseudomonas aeruginosa was 83% resistant for Amoxicillin, Cefotaxime, Ceftriaxone, Ciprofloxacin and Cetazidine 83%. Also resistance to Gentamycin was 67%, Levofloxacin was 50%, and Ofloxacin was 16.6 %.

Pathogens has been isolated 5 from 7 specimens from Thoracic surgery ward. The most common pathogen respectively was Klebsilla pneumonia 5 (100%), Enterobacter spp. was 3 (40%), pseudomonas putida 3(40%), staph. albus was 3(40%) and strepto.pyogens was 2(20%). as shown in table 2.

Table 2. Common pathogens isolated from thoracic surgery ward

Pathogen	Number	Percentage %
Klebsiella pneumniae	5	35.71
Enterobcter spp	3	21.43
pseudomonasputida	3	21.43
Staphylococcus albus	2	14.29
Streptococcus pyogenes	1	7.14
TOTAL	Total=14	100

The antimicrobial resistance profile of bacterial isolates showed that klebsilla Cefotaxime , ceftriaxone , gentamicin , imipenem , 100%, and Amoxicillin , ceftriaxone , ciprofloxacin 66.6%, and colistin , ofloxacin , Amikacin , 33.3%.

Pathogens has been isolated 4 from 3 specimens from Males surgical ward. The most common pathogen respectively were Acinetobacter Baumanni and Klebsilla pneumonia were 2 (50%), Citrobacter spp. And pseudomonas aeruginosa were 1(25%) shown in table 3.

Table 3. Common pathogens isolated from Males surgical ward

Pathogens	Number	Percentage %
Acinetobacter baumannii	2	33.3
Klebsiella spp	2	33.3
Citrobacter spp	1	16.7
Pseudomonas spp	1	16.7
Total	Total=6	100

The antimicrobial drug resistance profile of bacterial isolates showed that klebsilla was 50% resistant for Cefotaxime, imipenem, 50%, and Amoxicillin , cftazidime , ceftriaxone , ciprofloxacin, gentamicin , levofloxacin, 25%.

Pathogens has been isolated 4 from 3 specimens from Neurosurgery ward. The common pathogen were equals in percentage were Acinetobacter Baumanni and Klebsilla pneumonia, Ochrobacter anti horpi. And staph. Albus were 1 (25%). As shown in table 4

Table 4. Common pathogens isolated from Neurosurgery ward

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Pathogen	Number	Percentage %
Klebsiella pneumoniae	1	25
Ochrobacter antihropi	1	25
Acinetobacter baumannii	1	25
staphylococcus	1	25
TOTAL	TOTAL=4	100

The antimicrobial drug resistance profile of bacterial isolates showed that Ochrobacter Antihropi was 25%. klebsilla pneumonia was 25% and Acinetobacter baumanni was 25%. Staphylococcus albus was 25% resistant for Amoxicillin, Cefotaxime, ceftaadame, Ceftriaxone, colistin, gentamicin, imipenem, 100%.

Pathogens has been isolated 8 from 3 specimens from Females surgical ward. . The most common pathogen respectively was Escherichia Coli was 3(37.5%) Proteus mirabilis was 2(25%) .pathogen were equals in percentage were Bacillus spp and Klebsilla pneumonia, Staphylococcus aureus, Pseudomonas aeruginosa, Streptococcus pyogenes And Chryseomonas spp were 1 (25%) . as shown in table 5.

Table 5. Common pathogens isolated from Females surgery ward

PATHOGENS	Number	Percentage %
Escherichia Coli	3	27.3
Proteus mirabilis	2	18.18
Bacillus spp	1	9.1
Staphylococcus aureus	1	9.1
Klebsilla pneumoniae	1	9.1
Pseudomonas aeruginosa	1	9.1
Streptococcus pyogenes	1	9.1
Chryseomonas spp	1	9.1
TOTAL	TOTAL=11	100

The antimicrobial resistance profile of bacterial isolates showed that klebsilla pneumonia was 12.5%, Amoxicillin , Cefotaxime , cftazidime , ceftriaxone , Ofloxacin , ciprofloxacin , gentamicin , imipenem , levofloxacin , 100%.

Pathogens has been isolated 3 from 4 specimens from Males Orthopedics ward. The common pathogen were equals in percentage were Klebsilla pneumonia 3 (100%), Enterobacter spp.2 (66.6%) and Acinetobacter Baumanni 1 (33.3%). as shown in table 6.

Table 6. Common pathogens isolated from Males Orthopedics ward

Pathogens	Number	Percentage %
Klebsilla pneumoniae	3	50
Enterobacter cloacae	2	33.3
Acinetobacter baumanni	1	16.7
TOTAL	TOTAL=6	100

The antimicrobial drug resistance profile of bacterial isolates showed that Enterobacter cloacae was 66.6% resistant for Amoxicillin , ciprofxacin , gentamicin , imipenem , levofloxacin , 50%. Also Ampicillin, Cefotaxime, ceftaadame , ceftriaxone , colistin , 25%.
Pathogens has been isolated 6 from 4 specimens from out patients department. The most common pathogen respectively was Pseudomonas aeruginosa was 5(83.3%), The common pathogens were equals in percentage were Klebsilla pneumonia, Pantoea agglomerans , Staph. Aureus, Enterobacter sakazakii and E.coli 1 (16.6%). As shown in table 7.

Table 7. Common pathogens isolated from our patients department

Pathogens	Number	Percentage %
Pseudomonas aeruginosa	5	50%
Klebsilla pneumoniae	1	10%
Pantoea agglomerans	1	10%
Staph. Aureus	1	10%
Enterobacter sakazakii	1	10%
E.coli	1	10%
TOTAL	TOTAL=10	100%

The antimicrobial Drugs resistance profile of bacteria isolated 5 showed that pseudomonas aeruginosa was 83.3%, klebsilla pneumonia, Pantoea agglomerans, staphylococcus aureus, Enterobacter sakazakii, Escherichia coli was 16.6%. Pseudomonas: resistant for cftazidime was 100%, ceftriaxone was 60% imipenem was 80%.

4. DISCUSSION

Nosocomial infections, including surgical site infection, still form a large health problem and contribute substantially to patient morbidity, mortality, prolonged hospital stay, expensive hospitalization and prolonged therapy (Mulu et al., 2006; Sartelli et al., 2020). Emergence of poly antimicrobial resistant strains of hospital pathogens has also presented a major challenge in the provision of good quality in- patient care (Kamat et al., 2008). Antibiotics such as Ampicillin, Amoxicillin, Penicillin, Trimethoprim, Sulphamethoxazole, Chloramphenicol and Ceftriaxone are not the drug of choice for treating patients with nosocomial SSI in the study area where resistance to antibiotics reported high rates reached 100% in many situations.
The current study was incomparable with findings of another local studies in Libya conducted by Daw et al., 2023 (Tripoli Medical Center, Tripoli-Central Hospital, Benghazi Medical Center, and Sabha Medical Center) and Tajoury , 2019 (Benghazi), both studies reported S. aureus as the major causative bacterium of surgical wound infections reported were 42.6%, 19.9% respectively, also a Sudanese study reported S. aureus (55.0%)as a main cause of SSI, While in this study Klebsilla pneumonia and Acinetobacter baumanni were the most frequently detected bacterium (19%), S. aureus infection represented less than 5%. On the other hand, Mahjoub et al.(2013), found that Pseudomonas aeruginosa is the main causative bacterium of surgical infection in his study at Tripoli Medical Centre.
Our findings were also comparable with a review study reported the current state of antimicrobial resistance in Libya concluded that resistance to frequently administered antibiotics is very widespread in Libya (Rishi et al., 2013).

The predominance of Acinetobacter baumanni infection seen in this study especially in ICU and surgical ward is most likely associated with endogenous source as the organism is a member of the skin and nasal flora of the patients as it was explained by Isbori et al (2008). Infection with this organism may also be associated with contamination from the environment, surgical instruments or contaminated hands of the health professionals (Isbori et al., 2005 & Lilani et al., 2005).

5. CONCLUSION

The rate of nosocomial infection obtained in this study was comparable to other similar studies carried out in developing countries including Ethiopia. However, the bacterial isolates detected from our patients were terribly resistant for commonly available and prescribed antimicrobial drugs. Therefore, antibiotics such as Ampicillin, Amoxicillin, Penicillin, Trimethoprim, Sulphamethoxazole, Chloramphenicol and Ceftriaxone are not the drug of choice for treating patients with nosocomial infections in the study area. Al Jalla Referral Hospital also needs to make a concerted effort to minimize hospital acquired infections by following strict aseptic operation procedures, effective methods of sterilization and patient management.

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