Zagazig J. Pharm. Sci. June, 2019 ISSN 1110-5089
Vol. 28, Issue 1, pp. 13-24 ISSN (on-line) 2356_9786

Study on increased antimicrobial resistance among bacteria isolated from Intensive Care Units at Zagazig University Hospitals

Shaimaa I. Nazeih[1], * Fathy Serry[1], Hisham Abbas[1].
[1]*Department of Microbiology and immunology department, faculty of pharmacy, Zagazig university, Zagazig,Egypt.*
***Corresponding author:** E-mail:* shaimaa_nazeih@yahoo.com **,** *Tel/Fax: +201129844935*
Running title: Study on increased antimicrobial resistance.

Abstract

Emergence of antimicrobial resistance among the most important bacterial pathogens is recognized as a major public health threat affecting humans worldwide. Infections caused by resistant bacteria lead to up to two-fold higher rates of adverse outcomes compared with similar infections caused by susceptible strains. The negative impacts of antibacterial resistance can be measured at the patient level by increased morbidity and mortality, at the healthcare level by increased resource utilization, higher costs and reduced hospital activity and at the society level by antibiotic treatment guidelines favoring increasingly broad-spectrum empiric therapy.

In this study 67 isolates were collected from patients admitted to ICUs of Zagazig University Hospitals, Sharqia, Egypt. The isolates were biochemically identified and their susceptibility to different antimicrobials were tested by Kirby-Bauer standard disk diffusion method.

The results showed that the recovered bacteria had high degree of resistance to different antimicrobial classes and 86.25 % were multi drug resistance (MDR). In conclusion high rate of MDR were found in this study that necessitate strict antibiotic dispensing policy to reduce the increased antibiotic resistance.

Key words: Bacterial resistance, Antibiotics, MDR.

Introduction

Multidrug-resistant bacteria in both the hospital and community environment are of important concern to the clinician and the pharmaceutical industry, as it is the major cause of failure in the treatment of infectious diseases. Acquired antimicrobial resistance results in escalating healthcare costs, increased morbidity and mortality and the evolution of new pathogens **(Jones and Phaller, 1998)**.

During the last few decades the frequency and spectrum of antibiotic resistant infections have increased steadily within the United States, Europe and the developing world. This increase has been attributed to a combination of microbial characteristics, the selective pressure of antimicrobial use, and social and technical changes that enhance the transmission of resistant organisms factors, such as increased use and misuse of antimicrobial agents, increased use of invasive devices and

procedures, a greater number of susceptible hosts, and lapses in infection control practices leading to increased transmission of resistant organisms **(Harbarth *et al.*, 2001)**.

Microorganisms have a remarkable array of mechanisms with which to overcome the effects of antimicrobial agents. These include the production of structure-altering or inactivating enzymes (eg, beta-lactamases-or amino glycoside-modifying enzymes), alteration of penicillin-binding proteins or other cell-wall target sites, altered DNA gyrase targets, permeability mutations, active efflux and ribosomal modification **(Levy, 2002)**.

Selective pressure resulting from antimicrobial administration can lead to the growth of previously susceptible strains that have acquired resistance or to the overgrowth of strains that are intrinsically resistant. In general, resistance is acquired by mutational change or by the acquisition of resistance-encoding genetic material. The escape of resistance genes to mobile DNA fragments (plasmids) is enabling the process of transfer of antimicrobial resistance not only between bacteria of the same population, but also between bacteria from different genera. These evolutionary old genetic recombination mechanisms for gene transfer in bacteria have been adapted for new antibiotic environment that has been created due to liberal use of antibiotics in human medicine, agriculture, fisheries and animal husbandry **(Witte *et al.*, 1999)**.

In clinical practice, widespread use of antimicrobials in the intensive care units (ICUs) and for immunocompromised patients has resulted in the selection of multidrug-resistant organisms. Treatment of nosocomial infection caused by multidrug resistant microorganism is directly (increased infection control cost) and indirectly (prolonged hospital stay, increased laboratory cost) increasing health care cost **(Stone *et al.*, 2002)**.

Increased incidence of multidrug resistant bacteria and rising evidence of resistance transfer from one organism to another may lead to combined growth of nosocomial pathogens, for which there are no antibiotic solutions **(Jones and Phaller, 1998)**.

Isolates were completely identified and sensitivity patterns and MICs were determined. It is important to recognize that the concept of antimicrobial resistance/ susceptibility in clinical practice is a relative phenomenon with many layers of complexity. The establishment of clinical susceptibility breakpoints (susceptible, intermediate and resistant) mainly relies on the *in vitro* activity of an antibiotic against a sizeable bacterial sample, combined with some pharmacological parameters (e.g., blood and infection site concentrations of the antimicrobial, among others). Thus, when treating antibiotic-resistant bacteria, the interpretation of susceptibility patterns may vary according to the clinical scenario and the availability of treatment options.

This study aims to highlights the effect of misuse of chemotherapeutic antibiotics which leads to increase antibiotic resistance among pathogenic bacteria and becoming a rising problem for public health in recent decades.

Material and Methods

Bacterial strains

One hundred and three (103) Clinical specimens were obtained from patients in different surgical intensive care units (ICUs) of Zagazig University Hospitals, Sharqia, Egypt. The clinical samples were collected as tracheal aspirates, surgical wound swabs, blood, urine specimens, CVP and tracheoctomy swabs. All the specimens were collected aseptically and transported to the microbiology laboratory, Department of Microbiology and Immunology, Faculty of Pharmacy, Zagazig University where they were immediately processed and the bacterial pathogens were isolated and identified.

Some isolates were collected from culture collection department of Microbiology and Immunology, Faculty of Pharmacy, Zagazig University

Media and chemicals

Antibiotics disks were obtained from Oxoid, Hampshire, England. These disks included penicillin (P, 10 units), amoxicillin (Ax, 25 µg), amoxicillin-clavulanic acid (AMC, 20/10 µg), ampicillin (AM, 10 µg), ampicillin-sulbactam (SAM, 20 µg), methicillin (ME, 5 µg) piperacillin (PRL, 100 µg), piperacillin-tazobactam (PTZ, 110 µg), cefoperazone (CEP, 75 µg), cefepime (FEP, 30 µg), cefazolin (CZ, 30 µg), cefotaxime (CTX, 30 µg), ceftriaxone (CRO, 30 µg), ceftazidime (CAZ, 30 µg), cefoxitin (FOX, 30 µg), tetracycline (TE, 30µg), doxycycline (DO, 30 µg), tigecycline (TGC, 15 µg), nalidixic acid (NA, 30 µg), norfloxacin (NOR, 30 µg), gatifloxacin (GAT, 5 µg), ciprofloxacin (CIP, 5 µg), methicillin (ME, 5 µg), erythromycin (E, 15 µg), azithromycin (AZM, 15 µg), gentamicin (CN,10 µg), tobramycin (TOB, 10 µg), imipenem (IPM, 10 µg), meropenem (MEM, 10 µg), linzeolid (LZD, 30 µg), chloramephenicol (C, 30 µg), vancomycin (VA, 30 µg), teicoplanin (TPN, 30 µg), clindamycin (DA, 2 µg), quinupristin-dalfopristin (QDA, 15 µg), daptomycin (DAP, 30 µg), colistin (CT, 10 µg), and sulfamethoxazole-trimethoprim (SXT, 25 µg). The culture media that were used in this study included; Nutrient broth and agar, Muller Hinton broth and agar, MaCconkey's agar, Mannitol salt agar and that were obtained from Oxoid (Hampshire, England)

Isolation and Identification

The microbial isolates were collected from patients admitted to (ICUs) of Zagazig University Hospitals, Sharqia, Egypt by using sterile swabs. After collection, swabs were streaked onto the surface of each of nutrient agar, blood agar, Mannitol salt agar and MaCconkey's Agar plates then incubated at 37ºC for 24 hr **(winn and Koneman, 2006)**

The bacterial isolates were picked from agar plates and presumptively identified by Gram stain, colony morphology and biochemical characters according to standard microbiological techniques **(winn and Koneman, 2006).** These tests were catalase, oxidase, Coagulase, Hemolysis on blood agar, Mannitol fermentation, gelatin liquefication and pigmentation on nutrient agar were used for identification of Gram positive bacterial isolates

Indole production test, methyle red test, Vogus Prouskaur test , Citrate utilization , reaction on Triple sugar iron test and nitrate reduction test were used for identification of Gram negative bacterial isolates.

After bacterial identification all isolates were stored at -80ºC as 20% glycerol stocks.

Antimicrobial susceptibility testing of the bacterial isolates

The test was performed according to Clinical and Laboratory Standards Institute guidelines **(CLSI, 2017a)** using standard diffusion disk method.

Plates were examined and diameters of the complete inhibition zones were measured in mm, and interpreted according to **CLSI (2017b).**

Gram-positive isolates were tested against B-lactams (penicillin, ampicillin, ampicillin-sulbactam, methicillin, imipenem, cefazolin & cefotaxime), tetracyclines (tetracycline,)fluoroquinolones (ciprofloxacin), macrolides (erythromycin & azithromycin), aminoglycosides (gentamicin), oxazolidinones (linzeolid), phenicols (chloramphenicol), glycopeptides (vancomycin), lincosamides (clindamycin), and folate inhibitors (sulfamethoxazole-trimethoprim).

While, antimicrobial disks tested against Gram-negative bacteria were B-lactams (amoxicillin, amoxicillin-clavulanate, piperacillin, piperacillin-tazobactam, cefepime, cefoperazone, imipenem, meropenem, cefotaxime, ceftriaxone ceftazidime), tetracyclines (tetracycline), quinolones andfluoroquinolones (ciprofloxacin), aminoglycosides (gentamicin & tobramycin), phenicols (chloramphenicol), lipopeptides (colistin), and folate inhibitors (sulfamethoxazole-trimethoprim).

Results

1.Isolation and identification of bacteria from clinical specimens

From the collected 103 clinical specimens, 67 bacterial isolates were detected and were identified according the standard bacterial protocol. The type and number of the isolates are shown in table 1.

Table 1. Type and number of bacterial isolates

Name of Microorganism	Number of isolates
Pseudomonas aeruginosa (***P. aeruginosa)***	10
Acinetobacter baumannii ***(A. baumannii)***	10
Proteus vulgaris (***P. vulgaris)***	10
Proteus mirabilis ***(P. mirabilis)***	
Escherichia coli ***(E. coli)***	7
Klebsiella pneumoniae ***(K. pneumoniae)***	10
Staphylococcus aureus ***(S. aureus)***	10
Coagulase Negative Staphylococci ***(CoNS)***	10

2. Identification of Gram positive bacterial isolates

S. aureus isolates showed golden yellow colonies on nutrient agar. While, CoNS isolates showed white colonies on nutrient agar. Complete identification of *S. aureus* and CoNS based on their biochemical characteristics is shown in Table 2.

Table 2. Identification of Gram-positive isolates

Test	*S. aureus*	CoNS
Catalase	+	+
Oxidase	-	-
Coagulase	+	-
Hemolysis on blood agar	β-hemolysis	γ-non-hemolysis
Mannitol fermentation	+	-
Gelatin liquefaction	+	-
Pigmentation on nutrient agar	Golden yellow pigmentation	Off-white colonies

3. Identification of Gram-negative isolates

3.1. Identification of lactose fermenting isolates

E. coli isolates were identified as Gram-negative single rods with lactose fermenting colonies (rose pink colonies on MacConkey agar). *Klebsiella* isolates were identified as Gram-negative single rods with lactose fermenting colonies (pink mucoid colonies on MacConkey agar). Complete identification of lactose fermenting isolates based on their biochemical characteristics is shown in Table 3.

Table 3. Identification of Gram-negative lactose fermenting isolates

Test	*E. coli*	*K. pneumoniae*
IMViC*	++--	--++
Motility	Motile	Non-motile
TSI agar**	A/A+ gas	A/A+ gas
O/F test***	O^+/F^+	O^+/F^+
Growth on EMB agar****	Black colonies with greenish metallic sheen	Black mucoid colonies

IMViC: Indole Methyl red Voges-Proskauer Citrate utilization tests.
**TSI: Triple Sugar Iron, A: acidic, K: alkaline, butt/ slant reaction.
*** O/F test: Oxidation-fermentation test.
****EMB agar: Eosin-methylene blue agar.

3.2 Identification of non-lactose fermenting isolates

P. aeruginosa isolates were identified as Gram-negative single rods, showing greenish or reddish brown pigmentation on nutrient agar with characteristic grape juice-like odor. Meanwhile, *Proteus* isolates were identified according to their morphological appearance under microscope and the characteristic swarming and foul-like smell on nutrient agar. *A. baumannii* isolates were presumptively identified as Gram-negative cocco-bacilli with non-lactose fermenting colonies (slightly pinkish colonies on MacConkey agar). Complete identification of non-lactose fermenting isolates based on their biochemical characteristics is shown in Table(4)

Table 4. Identification of Gram-negative non-lactose fermenting isolates

Test	*P. aeruginosa*	*A. baumannii*	*P. mirabilis*	*P. vulgaris*
Oxidase	+	-	-	-
Catalase	+	+	+	+
IMViC*	- - - +	- - - +	- + - +	+ + - +
Urease	-	-	+	+
Motility	Motile	Non-motile	Motile	Motile
Swarming motility	-	-	+	+
TSI agar**	K/K	K/NC	K/A+ gas+ H_2S	K/A+ gas+ H_2S
O/F ***	O^+/F^-	O^+/F^-	O^+/F^+	O^+/F^+
Maltose fermentation	-	-	-	+
Growth at 44˚C	-	+	-	-
Arginine dihydrolase	+	-	-	-
Hemolysis on blood agar	β-hemolysis	γ- non hemolysis	γ- non hemolysis	γ- non hemolysis

*IMViC: Indole Methyl red Voges-Proskauer Citrate utilization tests
TSI: Triple Sugar Iron, A: acidic, K: alkaline, NC: no change. * O/F: Oxidation-fermentation.

Antimicrobial susceptibility testing: Determination of isolates susceptibility to different antimicrobials

The results showed that *Staphylococcus* isolates showed high resistance prevalence against penicillin, cefazolin, ampicillin, cefotaxime, ampicillin-sulbactam and erythromycin (Figure 1). Moreover, *S. aureus* isolates were resistant to tetracycline (84.4%), doxycycline, gentamicin (74.2% each), tobramycin (67.5%), ciprofloxacin, and norfloxacin (70% each). While, the prevalence of resistant isolates was observed to imipenem, azithromycin was 52.5% each, gatifloxacin and sulfamethoxazole-trimethoprim was 51.7% each. For CoNS, the prevalence of resistant isolates was as follows to tetracycline (47%), azithromycin (60%), gentamicin (54%), tobramycin (50%), ciprofloxacin (60%), norfloxacin (55%), gatifloxacin (40%), and sulfamethoxazole-trimethoprim (50%). *Staphylococcus* isolates showed low resistance prevalence to clindamycin, chloramphenicol. No isolates showed resistance to vancomycin, teicoplanin, tigecycline, linzeolid, quinupristin-dalfopristin and daptomycin (Figure 1).

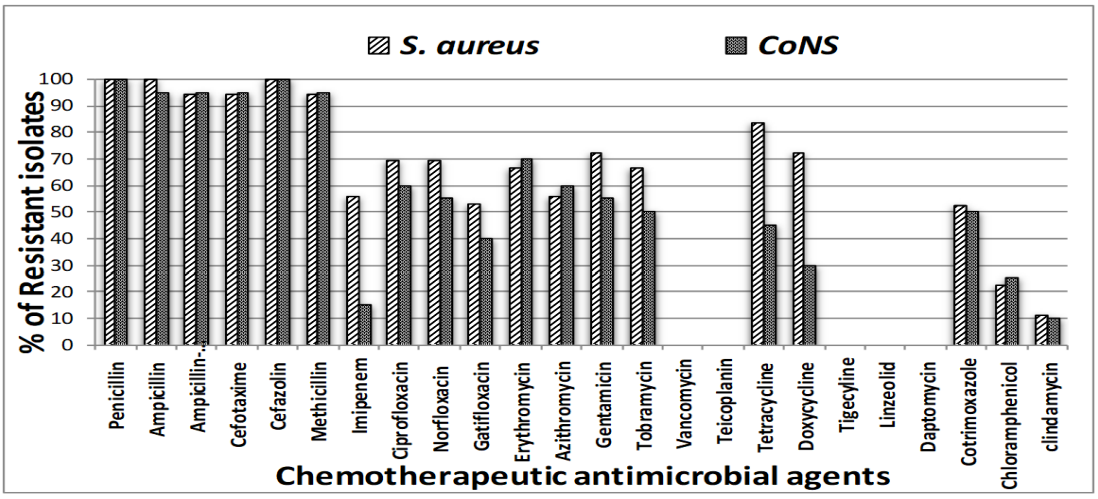

Figure 1. **Percentages of clinical *Staphylococcus* isolates resistant to various chemotherapeutic antimicrobial agents.**

Methicillin resistance (MRSA) was observed in 95.4% of *S. aureus* isolates and 96% of CoNS (MRCoNS) isolates **(Figure 1)**. Regarding Gram-negative isolates, *P. aeruginosa* and *A. baumannii* isolates were highly resistant to all tested antimicrobials except carbapenems and colistin **(Figure 2**). Furthermore, the resistance prevalence of *Klebsiella* spp. isolates was high to β-lactams, nalidixic acid, aminoglycosides and sulfamethoxazole -trimethoprim. *Klebsiella* spp. isolates showed intermediate resistance prevalence to piperacillin-tazobactam, tetracycline, doxycycline and chloramphenicol. In addition, *Proteus* spp. isolates showed high resistance to amoxicillin, amoxicillin-clavulanate, tetracyclines, aminoglycosides and intermediate resistance to piperacillin, cephalosporins and chloramphenicol.

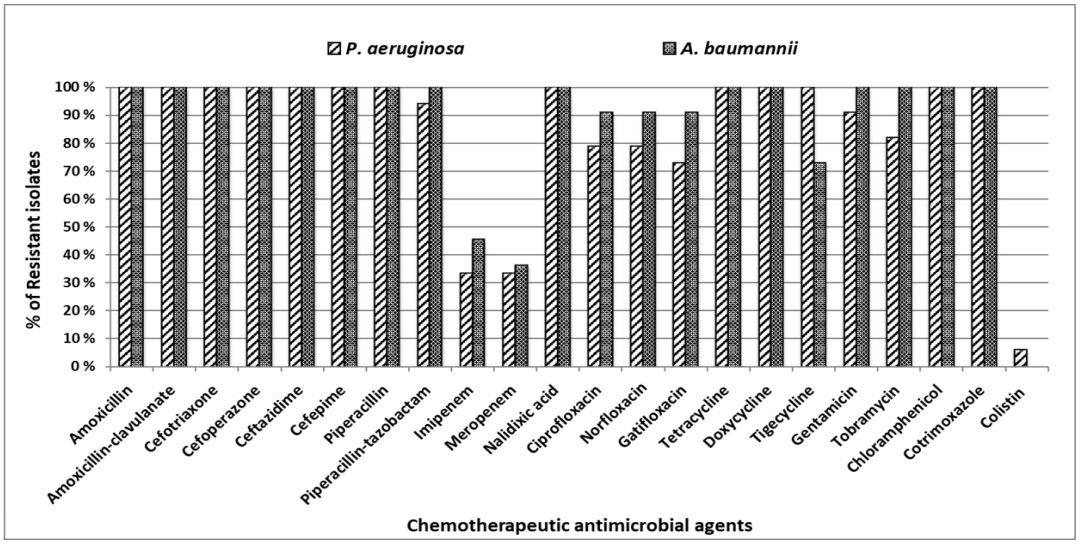

Figure 2. Percentages of clinical *P. aeruginosa* & *A. baumannii* resistant to various chemotherapeutic antimicrobial agents

.

Table 5. Percentages of clinical *Enterobacteriaceae* isolates resistant to various chemotherapeutic antimicrobial agents.

Antimicrobial agents	*Klebsiella* spp. (n=10)	*Proteus* spp. (n=20)	*E. coli* (n=7)
Amoxicillin	10 (100%)	19(95%)	7(100%)
Amoxicillin-clavulanate	10 (100%)	19(95%)	7(100%)
Ceftriaxone	10 (100%)	9(45%)	7(100%)
Cefoperazone	10 (100%)	11(55%)	7(100%)
Ceftazidime	10 (100%)	9(45%)	7(100%)
Cefepime	10 (100%)	9 (45%)	7(100%)
Piperacillin	7 (70%)	11(55%)	5(71.4%)
Piperacillin-tazobactam	6 (60%)	7(35%)	4(57.1%)
Imipenem	0 (0%)	3 (15%)	0 (0%)
Meropenem	0 (0%)	3 (15%)	0 (0%)
Nalidixic acid	8 (80%)	11(55%)	7(100%)
Ciprofloxacin	3 (30%)	5(25%)	5(71.4%)
Norfloxacin	3 (30%)	7(35%)	4(57.1%)
Gatifloxacin	1 (10%)	4(20%)	4(57.1%)
Tetracycline	5(50%)	20(100%)	7(100%)
Doxycycline	5(50%)	20(100%)	7(100%)
Tigecycline	0 (0%)	20(100%)	0 (0%)
Gentamicin	10 (100%)	15(75%)	7(100%)
Tobramycin	10 (100%)	16(80%)	7(100%)
Chloramphenicol	4 (40%)	13(65%)	4(57.1%)
Sulfamethoxazole-trimethoprim	7 (70%)	17(85%)	6(85.7%)

Discussion

Antibacterial therapy is one of the most important medical developments of the twentieth century; however, the spread of resistance in healthcare settings and in the community threatens the enormous gains made by the availability of antibiotic therapy. Infections caused by resistant bacteria lead to up to two-fold higher rates of adverse outcomes compared with similar infections caused by susceptible strains. These adverse outcomes may be clinical or economic and reflect primarily the failure or delay of antibiotic treatment. The magnitude of these adverse outcomes will be more pronounced as disease severity, strain virulence, or host vulnerability increases. The negative impacts of antibacterial resistance can be measured at the patient level by increased morbidity and mortality, at the healthcare level by increased resource utilization, higher costs and reduced hospital activity and at the society level by antibiotic treatment guidelines favouring increasingly broad-spectrum empiric therapy **(Friedman *et al.*, 2016).**

The resistance profile of all isolated bacteria was carried out by the Kirby-Bauer standard disk diffusion method according to **CLSI (2017b)** guidelines**.** Methcillin resistant *S. aureus* (MRSA) and Methicillin

resistant CoNs (MRCoNS) isolates constituted an alarmingly high percentage which was 95.4% and 96%, respectively **(Figure 1)**. These results are in accordance with that reported by **Song *et al.* (2001)** and **Ahmed *et al.* (2014)**. *Staphylococcus* isolates showed complete or high resistance to β-lactams, nalidixic acid, sulbactam-ampicillin, tetracycline, gentamicin, ciprofloxacin and erythromycin **(Figure 1)**. In accordance with these findings, the studies conducted by **Ahmad *et al.* (2013) and Perween *et al.* (2015)** explained the absolute resistance of *Staphylococcus* isolates to β-lactams and high resistance to gentamicin, ciprofloxacin and erythromycin. This study detected intermediate resistance prevalence of *S. aureus* to imipenem in agreement with **Elmanama *et al*. (2013)** who detected that 40% of *S. aureus* were resistant to imipenem. However, *Staphylococcus* isolates showed high susceptibility to clindamycin and chloramphenicol in accordance with **Saravanan *et al.* (2013).** No resistance was observed to vancomycin, tigecycline, linzeolid and daptomycin in agreement with **Mewara *et al.* (2014).**

Regarding Gram-negative isolates, *P. aeruginosa* isolates were completely resistant to amoxicillin, amoxicillin-clavulanate, all tested cephalosporins and tetracyclines **(Figure 2)**. A study reported by **Wang *et al.* (2012)** explained the absolute resistance of *P. aeruginosa* isolates to β-lactams which was in accordance with these results. This study is also supported by that of **Moazami-Goudarzi and Eftekhar (2013)** who reported high resistance to aminoglycosides, fluoroquinolones and piperacillin-tazobactam. The current study demonstrated that 34.6% *P. aeruginosa* isolates were resistant to carbapenems in agreement with **Mahmoud *et al.* (2013)**.

It was revealed that 46.4% of *A. baumannii* isolates were resistant to imipenem **(Figure 2)**. This result is conforming to data from 40 centers in 12 European countries participating in a monitory program which revealed that 42% of *A. baumannii* isolates were resistant to imipenem **(Turner, 2008)**. In agreement with the results recovered from this study, **Ziglam *et al.* (2012)** reported absolute resistance of *A. baumannii* isolated from Libyan BCU to β-lactams and aminoglycosides and high resistance to fluoroquinolones. This study showed that colistin was found to be the most active drug to both *A. baumannii* and *P. aeruginosa* in accordance with **Bayram *et al.* (2013)**.

In this study, the resistance of *Klebsiella* spp. isolates was complete to β-lactams and aminoglycosides in accordance with **Beheshti and Zia (2011)**. *Klebsiella* spp. isolates showed high resistance prevalence to sulfamethoxazole-trimethoprim, intermediate resistance prevalence to tetracycline, doxycycline and chloramphenicol **(Table 5)**. These findings are supported by **Sikarwar and Batra (2011)** who observed that *Klebsiella* spp. isolates were significantly resistant to piperacillin and sulfamethoxazole-trimethoprim and intermediately resistant to chloramphenicol and tetracycline. It was found that carbapenems and fluoroquinolones were the most effective antimicrobials against *Klebsiella* spp. in agreement with **Rao *et al.* (2014).** The present investigation revealed that *Proteus* spp. isolates showed high resistance prevalence to tetracyclines, aminoglycosides and penicillins in accordance with **Mordi and Momoh (2009)**. However, low

resistance rate was observed to piperacillin-tazobactam, fluoroquinolones and carbapenems in agreement with **Bhat and Vasaikar (2010).** *E. coli* isolates showed high resistance to all tested antibiotics except tigecycline and carbapenems **(Table 5)**. These findings are similar to the observation of **Ansari *et al.* (2015)**. The resistance of isolates was strikingly high. Multi drug resistant (MDR) was detected as resistance to at least one agent in three or more antimicrobial categories. It is revealed that 86.25% isolates were MDR isolates. All *P. aeruginosa, A. baumannii* and *E. coli* showed MDR. Moreover, MDR was observed in 93.6% of *Klebsiella* spp., 90% of CoNS, 75% of *S. aureus* and 73.3% of *Proteus* isolates. This is consistent with **Soleymanzadeh *et al.* (2013)** who reported that 88.43% of isolated pathogens were MDR isolates where they found that all *P. aeruginosa, A. baumannii,* and 93.75% of *Klebsiella* spp. isolates showed MDR. In addition, a study conducted by **Melake *et al.* (2015)** reported that 76.4% of *S. aureus* isolates were multidrug resistant staphylococci in agreement with this study. This striking high resistance may be recognized as the consequence of antibiotics misuse. Additionally, the reasons for this alarming phenomenon might be inappropriate and incorrect administration of antimicrobial agents in empiric therapies, prolonged hospitalization and lack of appropriate infection control strategies. All of these can cause a shift to increase prevalence of resistant organisms in the community **(Sosa *et al.*, 2010).** It is becoming increasingly documented that not only antibiotic resistance genes (ARGs) encountered in clinical pathogens are of relevance, but rather, all pathogenic, commensal as well as environmental bacteria—and also mobile genetic elements and bacteriophages—form a reservoir of ARGs (the resistome) from which pathogenic bacteria can acquire resistance via horizontal gene transfer (HGT). HGT has caused antibiotic resistance to spread from pathogenic bacteria to commensal and environmental species. Understanding the extent of the resistome and how its mobilization to pathogenic bacteria takes place is essential for efforts to control the dissemination of these genes **(Jones and Phaller, 1998)**.

Conclusion

Rapidly emerging resistant bacteria threaten the extraordinary health benefits that have been achieved with chemotherapeutics antibiotics. This crisis is global, reflecting the worldwide overuse of these antibiotics and the lack of development of new antibiotic agents by pharmaceutical companies to address the challenge. Antibiotic-resistant infections place a substantial health and economic burden on the health care system and population. Coordinated efforts to implement new policies, renew research efforts, and pursue steps to manage the crisis are greatly needed.

REFERANCES

Ahmad, B.; Urbas, F.; Johar Jamil, J. A.; Bashir, S. (2013). Biocides susceptibility pattern and phenotypic detection of Efflux pump in *Staphylococcus aureus* isolates from two tertiary hospitals of Pakistan. *African Journal of Microbiology Research, 7*(25): 3171-3178.

Ahmed, E. F.; Gad, G. F.; Abdalla, A. M.; Hasaneen, A. M.; Abdelwahab, S. F. (2014). Prevalence of methicillin resistant *Staphylococcus aureus* among Egyptian patients after surgical interventions. *Surg Infect (Larchmt), 15*(4): 404-411.

Ansari, S.; Nepal, H. P.; Gautam, R.; Shrestha, S.; Neopane, P.; Gurung, G.; Chapagain, M. L. (2015). Community acquired multi-drug resistant clinical isolates of *Escherichia coli* in a tertiary care center of Nepal. *Antimicrobial resistance and infection control, 4*(1): 1-8.

Bhat, V.; Vasaikar, S. (2010). Bacteriological profile and antibiogram of aerobic burn wound isolates in Mthatha, Eastern Cape, South Africa: original research. *Southern African Journal of Epidemiology and Infection, 25*(4): 16-19.

Bayram, Y.; Parlak, M.; Aypak, C.; Bayram, I. (2013). Three-year review of bacteriological profile and antibiogram of burn wound isolates in Van, Turkey. *International journal of medical sciences, 10*(1): 19-27.

Beheshti, S.; Zia, M. (2011). Bacteriology of burns and antibiogram in an Iranian burn care center. *African Journal of Pharmacy and Pharmacology, 5*(4): 538-541.

CLSI-Clinical and Laboratory Standards Institute (2017a). Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically: Approvated standard, CLSI Document M07-A9. Wayne, USA.

CLSI-Clinical and Laboratory Standards Institute (2017b). Performance standards for antimicrobial susceptibility testing, 17th informational supplement. CLSI Document M100-S23. Wayne, USA

Elmanama, A. A.; Al Laham, N. A.; Tayh, G. A. (2013). Antimicrobial susceptibility of bacterial isolates from burn units in Gaza. *Burns, 39*(8): 1612-1618.

Friedman, N.D.; Temkin, E.; Carmeli, Y. (2016). The negative impact of antibiotic resistance. *Clin Microbiol Infect.*(5):416-22.

Harbarth, S.; Albrich, W.; Goldmann, DA.; Huebner, J. (2001) Control of multiply resistant cocci: do international comparisons help? *Lancet Infect Dis*, 1: 251-61.

Jones, RN.; Phaller, MA. (1998). Bacterial resistance; a worldwide problem. *Diagn Microbiol Infect Dis*, 31: 379-88

Levy, SB.(2002) Active efflux, a common mechanism for biocide and antibiotic resistance. *J Appl Microbiol*, 92 Suppl: 65S-71S.

Mahmoud, A. B.; Zahran, W. A.; Hindawi, G. R.; Labib, A. Z.; Galal, R. (2013). Prevalence of multidrug-resistant

Pseudomonas aeruginosa in patients with nosocomial infections at a university hospital in Egypt, with special reference to typing methods. *Journal of Virology Microbiology, 2013*, 10-13.

Melake, N. A.; Eissa, N. A.; Keshk, T. F.; Sleem, A. S. (2015). Prevalence of multidrug-resistant bacteria isolated from patients with burn infection. *Menoufia Medical Journal, 28*(3): 677- 682.

Mewara, A.; Gautam, V.; Kaur, H.; Ray, P. (2014). *In vitro* evaluation of antibiotics for methicillin-resistant *Staphylococcus aureus* from north India. *The Indian journal of medical research, 139*(2): 319-327.

Mordi, R.; Momoh, M. (2009). Incidence of *Proteus* species in wound infections and their sensitivity pattern in the University of Benin Teaching Hospital. *African Journal of Biotechnology, 8*(5): 1-8.

Moazami-Goudarzi, S.; Eftekhar, F. (2013). Assessment of carbapenem susceptibility and multidrug-resistance in *Pseudomonas aeruginosa* burn isolates in Tehran. *Jundishapur Journal of Microbiology, 6*(2): 162-165.

Nannini, EC.; Singh, KV.; Arias, CA.; Murray, BE.; In vivo effect of cefazolin, daptomycin, and nafcillin in experimental endocarditis with a methicillin-susceptible Staphylococcus aureus strain showing an inoculum effect against cefazolin. *Antimicrob Agents Chemother. 2013;* 57(9):4276–81.

Perween, N.; Prakash, S. K.; Siddiqui, O. (2015). Multi Drug Resistant *Klebsiella* Isolates in Burn Patients: A Comparative Study. *Proteus, 71, 8*(4): 14-17.

Rao, S. P.; Rama, P. S.; Gurushanthappa, V.; Manipura, R.; Srinivasan, K. (2014). Extended-spectrum beta-lactamases producing *Escherichia coli* and *Klebsiella pneumoniae*: A Multi-centric Study Across Karnataka. *Journal of laboratory physicians, 6*(1): 7-16.

Saravanan, M.; Nanda, A.; Tesfaye, T. (2013). Antibiotic susceptibility pattern of methicillin resistant *Staphylococcus aureus* from septicemia suspected children in tertiary hospital in Hosur, South India. *American Journal of Microbiological Research, 1*(2): 21-24.

Sikarwar, A. S.; Batra, H. V. (2011). Prevalence of antimicrobial drug resistance of *Klebsiella pneumoniae* in India. *International Journal of Bioscience, Biochemistry and Bioinformatics, 1*(3): 211-215.

Soleymanzadeh, S.; Soltan Dallal, M.-M.; Amani, L.; Jabbari, M.; Rastegar Lari, A. (2013). Investigation of Resistant Bacteria in Nosocomial infections In Died Burn Patients during Hospitalization. *Infection, Epidemiology and Medicine, 1*(1): 19-25.

Song, W.; Lee, K. M.; Kang, H. J.; Shin, D. H.; Kim, D. K. (2001). Microbiologic aspects of predominant bacteria isolated from the burn patients in Korea. *Burns, 27*(2): 136-139.

Sosa, A. d. J.; Byarugaba, D. K.; Amabile-Cuevas, C. F.; Hsueh, P.-R.; Kariuki, S.; Okeke, I. N. (2010). Antimicrobial resistance in developing countries. New York, NY, USA: Springer.

Stone, PW,; Larson, E.; Kawar, LN. (2002) A systematic audit of economic evidence linking nosocomial infections and infection control interventions: 1990-2000. *Am J Infect Control*, 30: 145-52.

Turner, P. J. (2008). Meropenem activity against European isolates: report on the MYSTIC (meropenem yearly susceptibility test information collection) 2006 results. *Diagnostic microbiology and infectious disease*, *60*(2): 185-192.

Wang, L. J.; Sun, Y.; Song, W. L.; Zhang, Z. J.; Liu, C. F. (2012). Changes of drug-resistance of Pseudomonas aeruginosa in pediatric intensive care unit. *Zhonghua Er Ke Za Zhi*;50(9):657–663.

Witte, W.; Klare, I.; Werner, G.(1999) Selective pressure by antibiotics as feed additives. *Infection;* 27: S35-8.

Ziglam, H.; Elahmer, O.; Amri, S.; Shareef, F.; Grera, A.; Labeeb, M.; Zorgani, A. (2012). Antimicrobial resistance patterns among *Acinetobacter baumannii* isolated from burn intensive care unit in Tripoli, Libya. *The International Arabic Journal of Antimicrobial Agents*, *2*(3): 1-5.

دراسة عل زيادة المقاومة للمضادات العلاجية للبكتيريا المعزولة من وحدات العناية المركزة بمستشفيات جامعة الزقازيق

فتحي سري، هشام عباس، شيماء نزيه

قسم الميكروبيولوجي والمناعة، كلية الصيدلة، جامعة الزقازيق

يعد حدوث المقاومة للمضادات الحيوية ضمن اهم البكتريا الممرضة تهديدا حيويا للصحة العامة ويؤثر عل البشر في كل أنحاء العالم

وتسبب العدوي التي تحدثها البكتريا المقاومة لضعفين المشاكل مقارنة بالعدوي المشابهة التي تسببها البكتيريا الحساسية للمضادات الحيوية ويمكن قياس التأثير السلبي لزيادة المقاومة للمضادات العلاجية وذلك بزيادة الحالات المرضية وزيادة معدلات الوفاة وهذا يؤدي الي زيادة التكلفة العلاجية والفترة التي يقضيها المريض في المستشفي وبالتالي الاهدار من قدرة المستشفي علي استيعاب عدد اكبر من المرضي هذا علي مستوي المريض والمستشفي

اما بالنسبة للتأثير السيء لزيادة المقاومة للمضادات العلاجية عل المستوي المجتمعي فانه يؤدي الي الاضطرار الي اللجوء الي استعمال المضادات العلاجية واسعة المجال كحل سريع لعلاج العدوي البكتيريا

تستطيع البكتيريا ان تكون المقاومة للمضادات الحيوية عن طريق انتاج الإنزيمات المثبطة للمضادات الحيوية , زيادة نشاط مضخات التدفق , تقليل نفاذية الغشاء الخارجي للمضادات الحيوية و حدوث طفرات في مستقبلات المضادات الحيوية

في هذه الدراسة تم عزل ٦٧ عزلة من المرضي في وحدات العناية المركزة بمستشفيات جامعة الزقازيق بمحافظة الزقازيق في مصر وتم التعرف عليها باستخدام التجارب الكيمائية والحيوية وتم اجراء اختبار الحساسية للمضادات الميكروبية وقد أظهرت العزلات درجة عالية من تعدد المقاومة للمضادات الحيوية بنسبة ٨٦.٢٥٪

ويستنتج من هذه الدراسة ان هناك نسبة عالية من تعدد المقاومة للمضادات الحيوية مما يتطلب سياسة صارمة في صرف المضادات الحيوية من اجل تقليل ظاهرة المقاومة البكتريا للمضادات الحيوية