Influence of Different Families of Commercial Antibiotics on Controlling Nosocomial Infections

ABSTRACT

Aims: Nosocomial infection (NI) is one of the most important problems facing the world. This work is dedicated to investigating the prevalence rates of NI in addition to estimating the minimum inhibition concentration (MIC) of commercial antibiotics on isolated bacteria in order to determine the most diluted antibiotic that performed well with more efficiency and minimal toxicity.
Place and Duration of Study: Gastroenterology Surgical Center, Mansoura University, Mansoura, Egypt, between July 2017 and July 2018.
Methodology: This study included 368 different samples (urine, stool, sputum and surgical wounds) from 100 patients A total of 15 commercial antibiotics selected from seven families having different mode of action were used.
Results: Our findings demonstrated that the highest prevalence rate of NI was detected in *K. pneumonia*e (40%, n=56) followed by MRSA (22.85%, n=32), *E. coli* (20%, n=28), *P. mirabilis* (7.85%, n=11), *Ps. putida* (5%, n=7) and finally *Ps. aeruginosa* (4.3%, n=6). In addition, the results showed variable MICs of various antibiotics on isolated bacteria associated with NI.
Conclusion: Interestingly, our findings showed that quinolones family had the highest impact on all types of isolated bacteria associated with nosocomial infections.

Keywords: *Nosocomial infections, Antibiotics, K. pneumoniae, MRSA, E. coli, P. mirabilis, Ps. Putida, Ps. aeruginosa.*

INTRODUCTION

Nosocomial infection (NI) is considered to be the most difficult problems confronting clinicians who deal with severally ill patients. The incidence of NI is estimated at 5-10% in tertiary care hospitals reaching up to 28% in intensive care unit and 90% of these are caused by bacteria [1]. Generally, NIs can be defined as infection that were acquired during hospitalization by patients in whom the infection was not present or incubating at the time of admission [2, 3]. Additionally, NIs could be defined as any infections that were occurred within two days of hospital admission, three days of discharge or thirty days of an operation [4]. Indeed, NI could spread in the hospital and nursing home environments as well as clinic or other clinical settings. Interestingly, different routes for spreading NI to the susceptible patients in the clinical setting were reported. These routes include health care staff, contaminated equipment, bed linens and air droplets [5]. NIs could also be originated from the outside environment, another infected patient, or the patient's own skin microbiota, becoming opportunistic after surgery or other procedures that compromise the protective skin barrier [6]. On the other hand, antibiotic treatment is more effective when started early, as well as when it is adapted to the sensitivity of the pathogens[7]. In this work, we are concerning with assessing the influence of different families of commercial antibiotics, that were diluted serially, on the isolated bacteria obtained from cultured patients' samples who have NIs in order to determine the extent to which the isolated bacteria could be affected. This in turn may give a clue to determine the most diluted antibiotic that performed well with more efficiency and minimal toxicity.

MATERIALS AND METHODS

One hundred consecutive patients staying three days or more were randomly recruited from the Gastroenterology Surgical Center (GEC), Mansoura University, Egypt. Informed consents were obtained from all participants and they were fully informed concerning the diagnostic procedures involved and disease nature. The study protocol conformed to ethical guide-lines of 1975 Helsinki Declaration. Different samples of urine, stool, sputum and surgical wound swabs were collected from patients for microbiological studies in order to identify nosocomial infection. Next, the obtained samples were cultured using nutrient agar in addition to MacConkey agar. The isolated bacteria were then morphologically and biochemically identified by VITEK 2 compact 15 (Biomerieux, France) and in turn kept for subsequent analysis for assessing the influence of different commercial antibiotics on them. Interestingly, different families of commercial antibiotics (Table 1) were diluted serially from 10^{-1} to 10^{-5}. Next, a variety of cultured nutrient agar plates were then inoculated with 120 µL of each diluted antibiotic according to agar well-diffusion method[8].

RESULTS and DISCUSSION

No doubt, nosocomial infections are considered to be a serious public health issue. Of note, prevalence reports that were conducted in some developing countries have showed hospital-wide nosocomial infection rates mostly higher than 15% with a range from 6% to 27% [9].

The first part of this work was concerned with identifying different bacterial isolates related to nosocomial infections and estimating their prevalence rates in different samples. Three-hundred and sixty-eight samples were incorporated in this work. These samples were categorized into four different groups. They are urine (n=100), stool (n=100) and sputum (n=100) samples in addition to surgical wounds (n=68). These samples were further classified based on nosocomial infections into positive and negative groups as depicted in Figure 1A. Overall, one hundred and forty (38%) samples were tested positive for the presence of nosocomial infections bacteria.

On the contrary, sputum samples were accompanied with the lowest positivity rate of 27% for nosocomial infections as shown in Figure 1A lower than that reported previously by Custivic et al. [10] who showed that the nosocomial infections were detected in 60% in respiratory tract. On the contrary, our findings showed that 73.5% of surgical wounds, 28% of stool and 35% of urine samples were tested positive for the presence of nosocomial infections higher than that produced previously by Custivic et al. [10] who demonstrated that nosocomial infections were found in only 7%, 9% and 16% of surgical wounds, stool and urine samples, respectively.

It is noteworthy that nosocomial infections are caused by many microbes. However, bacteria are responsible for approximately 90% of nosocomial infections [11]. In this study, the agents that have been isolated from the included samples were *K. pneumoniae*, MRSA, *E. coli*, *P. mirabilis*, *Ps. putida* in addition to *Ps. aeruginosa*. Out of these bacteria, *K. pneumonia* and MRSA were found to have the highest detection rates. The results showed that *K. pneumoniae* had a prevalence rate in urine, stool and surgical wounds accounting for 40%, 46.43% and 42%, respectively. On the other hand, MRSA infection displayed the highest detection rate in sputum samples (51.9%).

Custivic et al. [10] reported that most MRSA infections occurred in the respiratory tract whereas MRSA accounted for 18.4% of all respiratory tract isolates. He also showed that *K. pneumoniae* was the most common, associated with more than half of surgical wound infections. On the other hand, Shaikh et al. [12] reported *E. coli* as the most frequent pathogen (26.3%) while *Klebsiella* spp. was responsible for only 5.2% of all urinary tract infections.

Overall, our results indicate that 25%, 20%, 19.3% and 35.7% of total positive samples of NIs was detected in urine, stool, sputum and surgical wound swaps samples, respectively. Based on the appearance of isolated bacteria in different samples, samples of

urine and surgical wounds swabs were found to be the best samples that could identify the bacterial nosocomial infections quickly. The positive samples were identified by VITEK 2 compact 15 (Biomerieux, France). As a consequence, different pathogenic bacteria were appeared with variable prevalence rates in different samples.

Overall, the present study showed that *K. pneumoniae* was found to have the highest detection rate of 40% (n=56) followed by MRSA (22.85%, n=32), *E. coli* (20%, n=28), *P. mirabilis* (7.85%, n=11), *Ps. putida* (5%, n=7) and finally *Ps. aeruginosa* (4.3%, n=6) as depicted in Figure 1B. However, Borg et al. [13] reported that the highest levels of MRSA were observed in Jordan, Egypt and Malta, which were all above 50% higher than that provided in this study. Custivic et al.[10] reported that *K. Pneumoniae* was the most frequently isolated gram negative pathogen (51% of all positive samples), followed by *Ps. aeruginosa* (10%), *P. mirabilis* (7%) and *E. coli* (4%), respectively; while MRSA was the most common among Gram positive bacteria (15%).

The second part of this work was dedicated to investigating the influence of different families of commercial antibiotics on nosocomial infections. Generally, antibiotics were selected from seven groups having different mode of action. They are quinolones, β-lactams, aminoglycosides, macrolides, tetracyclines, cephalosporines and glycopeptides. Thus, the efficiency of different families of commercial antibiotics in terms of minimum inhibition concentration (MIC) was then measured against the aforementioned bacteria associated with nosocomial infections and the results were summarized in Table 2 and Figure 2.

The obtained MIC for quinolones were found to be the same (10^{-4}) in case of *E. coli*, MRSA, *Ps. aeruginosa* and *P. mirabilis* and the maximum antibiotic activity was found in levofloxacin followed by ciprofloxacin. The same goes for β-lactam (MIC=10^{-4}) in case of *E. coli*, *P. mirabilis*, *Ps. aeruginosa* and *P. mirabilis* followed by MRSA (MIC=10^{-2}) and the maximum antibiotic activity was found in meropenem. The obtained MIC for aminoglycosides was found to be 10^{-5} and 10^{-4} in case of *E. coli* and *Ps. aeruginosa*, respectively, and the maximum antibiotic activity was found in amikacin (MIC=10^{-5}) with *E. coli* compared to other antibiotics. The obtained MIC for macrolides was found to be 10^{-4} and 10^{-2} in case of *E. coli* and MRSA, respectively. The obtained MIC for tetracycline was found to be 10^{-4} in case of MRSA and 10^{-2} in case of *P. mirabilis* and *Ps. aeruginosa*. Surprisingly, cephalosporin family showed no activity upon dilutions while its antibiotic activity was shown in case of its original form in *K. pneumoniae*, MRSA and *P. mirabilis*. Finally, the obtained MIC for glycopeptides was found to be 10^{-3} in case of MRSA and 10^{-1} in case of *E. coli*, *Ps. aeruginosa* and *Ps. putida*. Based on the aforementioned results, that quinolones family had the highest impact on all types of isolated bacteria associated with nosocomial infections.

Among these, the study that was reported by Lombardi et al. [14] who showed that *Ps. putida* was resistant to quinolones but it was susceptible to aminoglycosides except for amikacin. As well, Kim et al. [15] reported that *Ps. putida* was susceptible to ciprofloxacin, levofloxacin, amikacin and meropenem. Another study was reported by Ayub et al. [16] showed that *S. aureus* and *P. mirabilis* were resistant to amoxicillin but susceptible to tetracycline. In addition, Mustafa et al. [17] reported that MRSA was resistant only to β-lactams while *S. aureus* was resistant to penicillin and susceptible to nitrofurantoin, vancomycin, levofloxacin, tetracycline, and erythromycin. Mustafa et al. [17] also reported that the most potent antimicrobials on *E. coli* were meropenem, levofloxacin and amikacin. It was also reported that *K. Pneumoniae* was susceptible to amikacin and moderately sensitive to ceftriaxone, ciprofloxacin and tetracycline. Additionally, Mustafa et al. [17] reported that the most potent antimicrobials on *Ps. aeruginosa* were meropenem and levofloxacin while *Ps. aeruginosa* was resistant to tetracycline, erythromycin and vancomycin. This is comparable with the rates reported by various authors Maehara et al. [18], Aga et al. [19] and Baker et al. [20]

Table 1. The mode of action of common antibiotics

Antibiotic family	Scientific name	Abbreviation	Commercial name	Conc. (mg)	Mode of action
Quinolones	Ciprofloxacin	CIP	Ciprofloxacin	250	DNA Inhibitors
Quinolones	Norfloxacin	NOR	Noracin	400	DNA Inhibitors
Quinolones	Ofloxacin	OFX	Tarivid	200	DNA Inhibitors
Quinolones	Levofloxacin	LEV	Lee-Flox	250	DNA Inhibitors
Quinolones	Nitrofurantoin	F	Macrofuran	50	DNA Inhibitors
β-lactam	Meropenem	MEM	Meronem	1000	Cell wall Inhibitors
β-lactam	Cefotaxime	CTX	Cefotax	250	Cell wall Inhibitors
β-lactam	Amoxicillin	AX	E-Mox	1000	Cell wall Inhibitors
β-lactam	Ampicillin	AM	Epicocillin	500	Cell wall Inhibitors
Aminoglycosides	Amikacin	AK	Emijectacin	500	Protein Inhibitors
Aminoglycosides	Neomycin	N	Neomycin	500	Protien Inhibitors
Macrolides	Erythromycin/Trimethoprim	E	Erythroprim	400/100	Protien Inhibitors
Tetracyclines	Tetracycline	TET	Cefaxone	250	Protien Inhibitors
Cephalosporines	Ceftriaxone	CRO	Doxymycin	100	Cell wall Inhibitors
Glycopeptides	Vancomycin	V	Vancomycin	500	Cell wall Inhibitors

Table 2. Minimum inhibition concentration (MIC) of different families of antibiotics on bacterial isolates associated with nosocomial infections

Family[a]	Abbreviation[b]	MIC					
		E. coli	*K. pneumoniae*	*P. mirabilis*	MRSA	*Ps. aeruginosa*	*Ps. putida*
F1	CIP	$1x10^{-4}$	$1x10^{-1}$	$1x10^{-4}$	$1x10^{-4}$	$1x10^{-4}$	$1x10^{-3}$
	NOR	$1x10^{-4}$	$1x10^{-1}$	$1x10^{-4}$	$1x10^{-4}$	$1x10^{-3}$	$1x10^{-2}$
	OFX	$1x10^{-4}$	$1x10^{-1}$	$1x10^{-4}$	$1x10^{-4}$	$1x10^{-2}$	$1x10^{-2}$
	LEV	$1x10^{-4}$	$1x10^{-2}$	$1x10^{-4}$	$1x10^{-4}$	$1x10^{-4}$	$1x10^{-3}$
	F	$1x10^{-2}$	1	1	1	1	1
F2	MEM	$1x10^{-4}$	$1x10^{-1}$	$1x10^{-4}$	$1x10^{-2}$	$1x10^{-4}$	$1x10^{-4}$
	CTX	$1x10^{-4}$	1	1	$1x10^{-1}$	$1x10^{-2}$	$1x10^{-2}$
	AX	$1x10^{-4}$	1	$1x10^{-1}$	$1x10^{-2}$	$1x10^{-1}$	$1x10^{-1}$
	AM	$1x10^{-4}$	1	$1x10^{-1}$	$1x10^{-2}$	$1x10^{-1}$	$1x10^{-1}$
F3	AK	$1x10^{-5}$	$1x10^{-2}$	$1x10^{-2}$	$1x10^{-3}$	$1x10^{-4}$	$1x10^{-3}$
	N	$1x10^{-2}$	$1x10^{-1}$	$1x10^{-2}$	$1x10^{-2}$	$1x10^{-3}$	$1x10^{-3}$
F4	E	$1x10^{-4}$	1	$1x10^{-1}$	$1x10^{-2}$	$1x10^{-1}$	1
F5	TET	$1x10^{-1}$	$1x10^{-1}$	$1x10^{-2}$	$1x10^{-4}$	$1x10^{-2}$	$1x10^{-1}$
F6	CRO	$1x10^{-4}$	1	1	1	$1x10^{-2}$	$1x10^{-3}$
F7	V	$1x10^{-1}$	1	1	$1x10^{-3}$	$1x10^{-1}$	$1x10^{-1}$

[a] F1: Quinolones; F2: β- lactams; F3: Aminoglycosides; F4: Macrolides; F5: Tetracyclines; F6: Cephalosporines and F7: Glycopeptides. [b] Show Table 1.

Figure 1. Different bacterial isolates associated with nosocomial infections (A) Prevalence rate of bacterial isolates in different patient samples, and (B) Types of isolated bacteria and their detection rate in different samples.

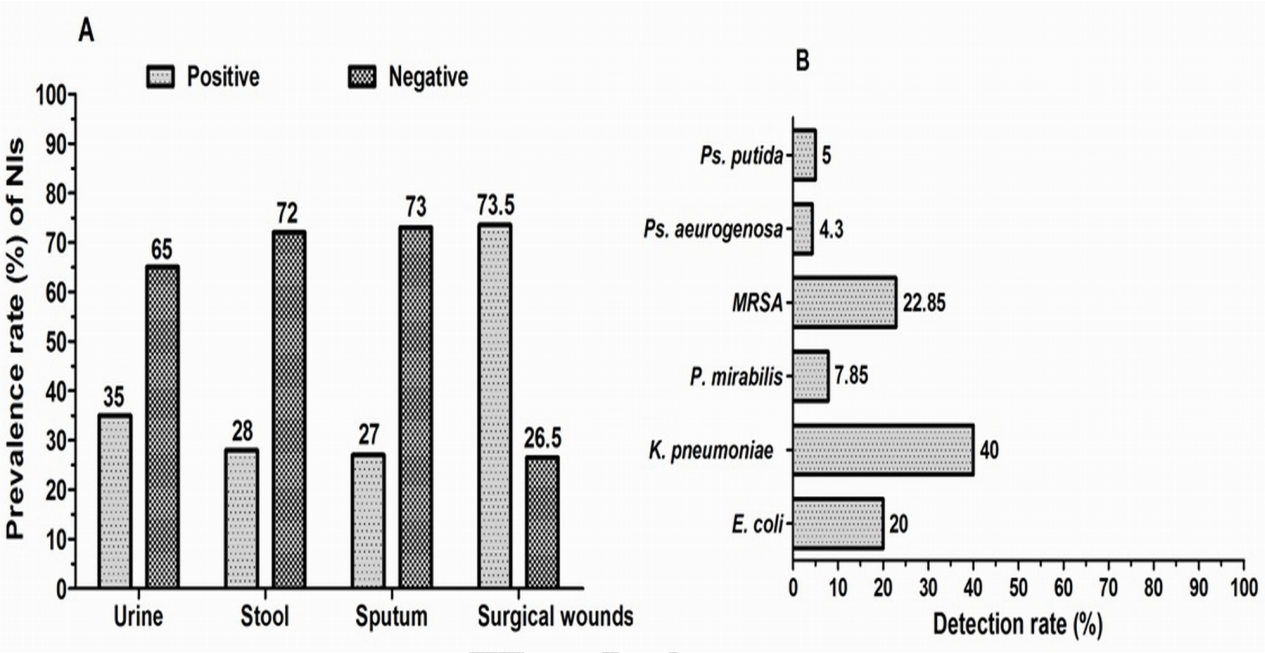
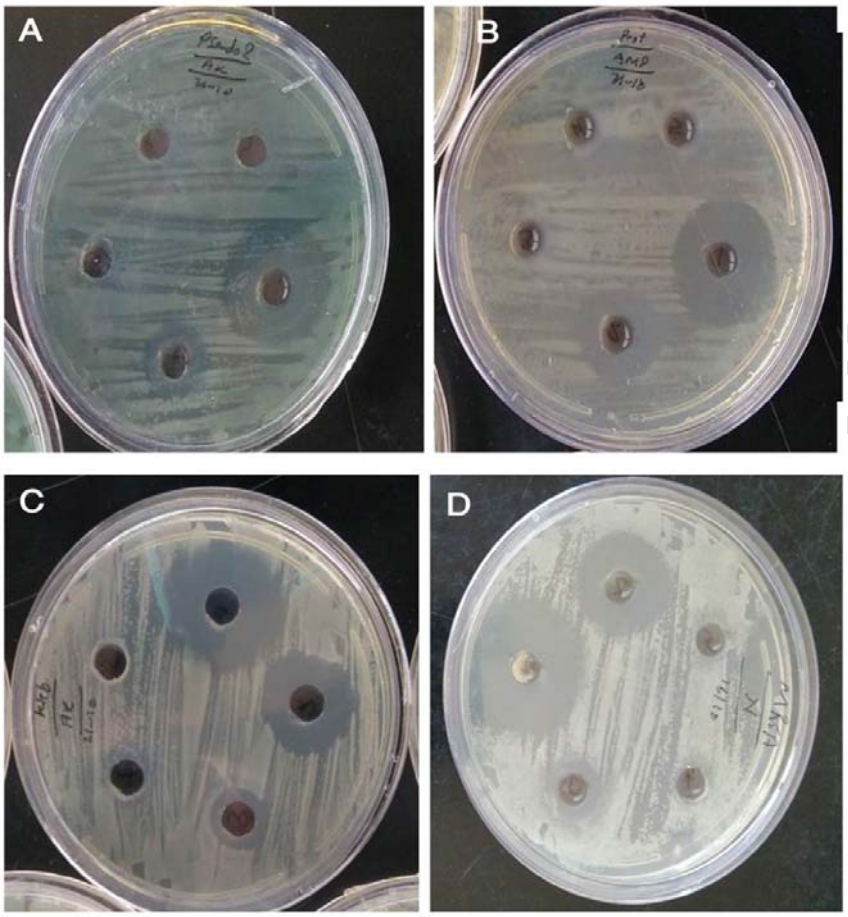


Figure 2. Influence of some commercial antibiotics with different concentration on isolated bacteria associated with nosocomial infections (A) Amoxicillin on *Ps. aeruginosa*, (B) Ampicillin on *P. mirabilis*, (C) Amikacin on *K. pneumoniae*, and (D) Neomycin on MRSA.



CONCLUSION

Our findings showed that quinolones family had the highest impact on all types of isolated bacteria associated with nosocomial infections.

REFERENCES

1. Jain A, Singh K. Recent advances in the management of nosocomial infections. JK Science. 2007;9(1):3-8.
2. Struelens MJ. The epidemiology of antimicrobial resistance in hospital acquired infections: problems and possible solutions. Bmj. 1998;317(7159):652-4.
3. Ducel G, Fabry J, Nicolle L, Organization WH. Prevention of hospital-acquired infections: a practical guide. 2002.
4. Inweregbu K, Dave J, Pittard A. Nosocomial infections. Continuing Education in Anaesthesia, Critical Care & Pain. 2005;5(1):14-7.
5. Siegel JD, Rhinehart E, Jackson M, Chiarello L. Management of multidrug-resistant organisms in health care settings, 2006. American journal of infection control. 2007;35(10):S165-S93.
6. Dancer SJ. Importance of the environment in meticillin-resistant Staphylococcus aureus acquisition: the case for hospital cleaning. The Lancet infectious diseases. 2008;8(2):101-13.
7. Kumar A, Roberts D, Wood KE, Light B, Parrillo JE, Sharma S, et al. Duration of hypotension before initiation of effective antimicrobial therapy is the critical determinant of survival in human septic shock. Critical care medicine. 2006;34(6):1589-96.
8. Valgas C, Souza SMd, Smânia EF, Smânia Jr A. Screening methods to determine antibacterial activity of natural products. Brazilian journal of microbiology. 2007;38(2):369-80.
9. Pittet D, Allegranzi B, Storr J, Nejad SB, Dziekan G, Leotsakos A, et al. Infection control as a major World Health Organization priority for developing countries. Journal of Hospital Infection. 2008;68(4):285-92.
10. Custovic A, Smajlovic J, Hadzic S, Ahmetagic S, Tihic N, Hadzagic H. Epidemiological surveillance of bacterial nosocomial infections in the surgical intensive care unit. Materia socio-medica. 2014;26(1):7.
11. Khan HA, Ahmad A, Mehboob R. Nosocomial infections and their control strategies. Asian pacific journal of tropical biomedicine. 2015;5(7):509-14.
12. Shaikh JM, Devrajani BR, Shah S, Akhund T, Bibi I. Frequency, pattern and etiology of nosocomial infection in intensive care unit: an experience at a tertiary care hospital. J Ayub Med Coll Abbottabad. 2008;20(4):37-40.
13. Borg MA, De Kraker M, Scicluna E, van de Sande-Bruinsma N, Tiemersma E, Monen J, et al. Prevalence of methicillin-resistant Staphylococcus aureus (MRSA) in invasive isolates from southern and eastern Mediterranean countries. Journal of Antimicrobial Chemotherapy. 2007;60(6):1310-5.
14. Lombardi G, Luzzaro F, Docquier J-D, Riccio ML, Perilli M, Colì A, et al. Nosocomial infections caused by multidrug-resistant isolates of Pseudomonas putida producing VIM-1 metallo-β-lactamase. Journal of clinical microbiology. 2002;40(11):4051-5.
15. Kim SE, Park S-H, Park HB, Park K-H, Kim S-H, Jung S-I, et al. Nosocomial Pseudomonas putida bacteremia: high rates of carbapenem resistance and mortality. Chonnam medical journal. 2012;48(2):91-5.
16. AYUB S, FATIMA B, BAQIR S, NAQVI S, SHEIKH D, ALI SM, et al. AMOXICILLIN AND TETRACYCLINE ACTIVITY AGAINST STAPHYLOCOCCUS AUREUS AND PROTEUS MIRABILIS. Int J Curr Pharm Res.7(4):49-52.
17. Mustafa M-H, Khandekar S, Tunney MM, Elborn JS, Kahl BC, Denis O, et al. Acquired resistance to macrolides in Pseudomonas aeruginosa from cystic fibrosis patients. European Respiratory Journal. 2017;49(5):1601847.
18. Maehara Y, Shirabe K, Kohnoe S, Emi Y, Oki E, Kakeji Y, et al. Impact of intra-abdominal absorbable sutures on surgical site infection in gastrointestinal and hepato-biliary-pancreatic surgery: results of a multicenter, randomized, prospective, phase II clinical trial. Surgery today. 2017;47(9):1060-71.
19. Aga E, Keinan-Boker L, Eithan A, Mais T, Rabinovich A, Nassar F. Surgical site infections after abdominal surgery: incidence and risk factors. A prospective cohort study. Infectious Diseases. 2015;47(11):761-7.
20. Baker AW, Dicks KV, Durkin MJ, Weber DJ, Lewis SS, Moehring RW, et al. Epidemiology of surgical site infection in a community hospital network. infection control & hospital epidemiology. 2016;37(5):519-26.