

Evaluation of Bacterial Resistance to Antiseptics and Disinfectants in Clinical Isolates

Luma N. Abdulrazaq

Master degree in Pathology, Anbar Health Department Heet general hospital Anbar _Iraq

Ghada Nasrat Khalaf

Baghdad Health Directorate/Al-Karkh. Al-Doura Primary Health Care Sector

Ahmed Nafia Obaid

Hit General Hospital Anbar Health Department Anbar, Iraq

Mayyadah Ali Ahmed Al-Gburi

Salah al-Din Health Department Baiji Sector

Abdulsalam Abdulsattar Abdulazez

Medical Laboratory Techniques department, College of Health and medical technology, University of Al-maarif, Anbar, Iraq

Received: 2025, 15, Nov

Accepted: 2025, 21, Dec

Published: 2026, 19, Jan

Copyright © 2026 by author(s) and Scientific Research Publishing Inc. This work is licensed under the Creative Commons Attribution International License (CC BY 4.0).



Open Access

<http://creativecommons.org/licenses/by/4.0/>

Annotation: In this study 100 samples are collected from July 2021 to November 2021 from many hospitals in Anbar (Heet, Al- Ramadi and Haditha hospital). 59 sample isolates of *Pseudomonas aeruginosa* (*P. aeruginosa*) have been diagnosed from cases of infections of wounds and burns. There was different level of antibiotics resistance among bacterial isolates, the highest level of resistance was with meropenem 58 isolates (98.3%), followed by imipenem 52 isolates (88.1%), tobramycin and ofloxacin 51 isolates (86.4%), gentamycin 44 isolates (74.6%), ciprofloxacin 42 isolates (71.2%), amikacin 35 isolates (59.3%), ceftazidime 34 isolates (57.6%), cefotaxime 28 isolates (47.5%), and the lowest level of resistance was shown with ampicillin 16 isolates (27.1%). When studying the ability of *P.*

aeruginosa isolates to secrete biofilm, as one of the virulence factors, was found all of these isolates (100%). During this study, three types of disinfectants were used as chemical agents that have a role such as anti-biofilm, and the lowest concentration used to inhibit biofilm production by bacterial isolates was measured. MICs were determined as 0.0031, 0.0062 and 0.0125 for povidone-iodine, IPC231 and surfaxn, respectively.

Introduction

In most nations, disinfectants are widely utilized in the food processing, agricultural, healthcare, and domestic industries, cosmetics, and pharmaceuticals must adhere to stringent quality and safety regulations.[1]

Disinfectants and increased biosecurity are now the best line of defense against the spread of microorganisms in industrial and medical settings.[2]

Pseudomonas aeruginosa (*P. aeruginosa*) infections have occasionally been traced back to tainted medical equipment.[3]

Research has linked *P. aeruginosa* to a number of different human diseases. Opportunistic pathogens are known to occasionally infect healthy hosts but can cause life-threatening infections in immuno compromised individuals.[4]

Although *P. aeruginosa* seldom infects healthy tissues, it can cause a wide variety of infections, this includes the host's epithelial barriers, mucociliary layer, cystic fibrosis, a wide variety of systemic disorders, including urinary tract infections, respiratory infections, dermatitis, bacteremia, bone and joint infections, gastrointestinal infections [5]

Pseudomonas aeruginosa is the fourth most commonly isolated nosocomial bacteria, responsible for 10.1% of all hospital-acquired infections in the United States, according to the Centers for Disease Control and Prevention (CDC) [6].

Aim of study

The goal of this research was to use a variety of techniques to isolate bacteria from different sources and identify them, as well as to examine biofilm development utilizing these bacterial isolates. The disk diffusion method for testing bacterial susceptibility.

Material and Method

Sample collection

In this study 100 samples are collect from July 2021 to November 2021 from Heet, Al-Ramadi, Haditha hospitals in Anbar. These swabs are collected from hospitals workers (tool swabs) and the patients with burning and open wounds. About 59 isolates of *P. aeruginosa* have been diagnosed from different cases of infections.

Isolation and diagnosis of *P. aeruginosa*

After the collection the samples were inoculated on BA and MacConkey agar (MCA) for primary isolation. After the good pure grown colonies were isolated, on specific selective media Muller Hinton agar (MHA), Brain-Heart infusion broth (BHIB), Brain-Heart infusion agar (BHIA), and NA used and biochemical examines catalase enzyme, urease and for further identification, diagnosis and characterization were made.

The morphology of bacterial isolates grown in MCA medium was employed to make the diagnosis. This was evidenced by the lack of coloration and lactose non-fermentation in the bacterial colonies [7].

Antibiotic discs test

Amikacin, ciprofloxacin, imipenem, ceftazidime, cefotaxime, gentamicin, meropenem, ofloxacin, tobramycin, and ampicillin. Adjustments to the Bauer Kirby technique have been made in light of the results to produce a generally accepted standard [8].

Detection of some virulence factors

To make hemolysin, new bacterial isolates were grown on BA and incubated for 24 hours at 37 °C. Determination of *P. aeruginosa* virulence factors is successful if the colony contains hemolysin. Micro titer plate assays for biofilm generation can be utilized to identify virulence factors[9].

Results and discussion

Antibiotic Resistance Profile Among Bacterial Isolates

A wide range of antibiotic resistance was seen among the bacterial isolates tested in this investigation, with meropenem showing the highest rate of resistance (58 isolates, 98.3%), followed by imipenem (52 isolates, 88.1%), tobramycin and ofloxacin (51 isolates, 86.4%), gentamycin (44 isolates, 74.6%), ciprofloxacin (42 isolates, 71.2%), amikacin (35 individuals Antibiotic resistance, depicted in Figure 1, and described Table 1 below.



Figure 1: Antibiotic resistance for isolates of *P. aeruginosa* bacteria

Table 1: Antibiotic resistance distribution among bacterial isolates

Antibiotics	Sensitive	Resistance	MIS
Amikacin	22 (37.3%)	35 (59.3%)	2 (3.4%)
Ceftazidime	22 (37.3%)	34 (57.6%)	3 (5.1%)
Cefotaxime	30 (50.8%)	28 (47.5%)	1 (1.7%)
Ciprofloxacin	11 (18.6%)	42 (71.2%)	6 (10.2%)
Gentamycin	13 (22%)	44 (74.6%)	2 (3.4%)
Imipenem	1 (1.7%)	52 (88.1%)	6 (10.2%)
Meropenem	0 (0%)	58 (98.3%)	1 (1.7%)
Ofloxacin	4 (6.8%)	51 (86.4%)	4 (6.8%)

Ampicillin	36 (61.0%)	16 (27.1%)	7 (11.9%)
Tobramycin	7 (11.9%)	51 (86.4%)	1 (1.7%)

MIC of Disinfectants

In this work, the minimum dosage required to prevent biofilm generation by bacterial isolates was determined using three different types of disinfectants as chemical agents with roles such as anti-biofilm. As can be seen in Table 2 below, the minimum inhibitory concentration (MIC) for povidone-iodine was 0.0031, the MIC for IPC231 was 0.0062, and the MIC for surfaxn was 0.0125.

Table 2: MIC of disinfectants

Type of disinfectant used	Minimum inhibitory concentration (mg/ml)	Certified concentration
Povidone-iodine	0.0031	10%
IPC231	0.0062	5%
Surfaxn	0.0125	20%

MIC of anti-biofilm disinfectants

Tables 3, 4, 5 and 6 detail the results of this study, in which the MIC of disinfectants was determined, as well as the extent of their effect on groups of bacterial isolates producing biofilm at different concentrations and times using CV and MTT biofilm assay.

Povidone-antibiofilm iodine's activity is laid bare in Tables 3, 4, 5 and 6. Strong biofilm producer strains of *Pseudomonas* were shown to be considerably suppressed ($P \leq 0.01$) by MIC values, shifting the phenotypic profile to that of a moderate biofilm producer, as seen by the CV and MTT stain, assay. Although there was no noticeable shift in phenotype, the weak biofilm producer strain *Pseudomonas* was likewise strongly suppressed by the povidon concentrations evaluated.

Even after applying povidone-iodine to preexisting biofilms for an hour, we were unable to confirm that the biofilms had been completely removed. Exposure to MIC levels of povidone-iodine still resulted in a significant ($P \leq 0.01$) decrease in the biofilm of both the strong and moderate biofilm producers *Pseudomonas*.

Tables 3, 4, 5 and 6 show that IPC has antibiofilm activity. The phenotypic profile of *Pseudomonas*, a strong biofilm producer strain, was altered to that of a moderate biofilm producer after MIC values dramatically decreased biofilm formation ($P \leq 0.01$). *Pseudomonas*, a low-level biofilm producer, was also strongly suppressed by the tested amounts of IPC, although showing no alterations to its phenotypic profile.

After applying IPC to preexisting biofilms for 2 hours, we were unable to confirm that any of the biofilms had been completely removed. Still, the biofilm produced by the strong and moderate biofilm producers was drastically cut ($P \leq 0.01$). *Pseudomonas aeruginosa*, after being dosed with IPC at concentrations above the minimum inhibitory.

Table 3 MIC of anti-biofilm disinfectants-CV

Weak	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	P-value
Povidone-iodine	28.4	16.6	9.4	6.1	0.0001
IPC	29.5	17.4	10.2	8.3	0.0001
SURFAXN	29.5	17.5	10.7	8.9	0.0001
Moderate	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	-

Povidone-iodine	25.25	14.5	7.7	4.5	0.0002
IPC	26.5	15.75	8.9	5.3	0.0001
SURFAXN	26.3	15.75	8.6	5.6	0.0001
Strong	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	-
Povidone-iodine	16.5	11.5	6.2	2.7	0.0035
IPC	18.4	12.4	7.8	3.2	0.0044
SURFAXN	18.8	14.5	7.9	3.5	0.0037
<i>S. aureus</i>	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	-
Povidone-iodine	26.5	15.9	9.2	3.5	0.0019
IPC	26.5	15.45	8.9	4.3	0.0009
SURFAXN	26.375	15.75	8.6	5.6	0.0008
<i>E. coli</i>	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	-
Povidone-iodine	24.95	13.9	6.7	3.8	0.0013
IPC	25.5	14.7	8.9	4.5	0.0011
SURFAXN	26.8	14.8	8.6	4.8	0.0014

Table 4 MIC of anti-biofilm disinfectants-CV after 8 hours

Weak	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	P-value
Povidone-iodine	36.1	22.2	12.8	5.8	0.0033 **
IPC	36.8	23.4	13.1	6.2	0.0028 **
SURFAXN	37.6	24.3	14.2	6.5	0.0021 **
Moderate	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	---
Povidone-iodine	27.5	16.1	9.1	5.25	0.0052 **
IPC	27.2	16.5	9.8	6.1	0.0054 **
SURFAXN	29.3	16.5	9.7	6.375	0.0020 **
Strong	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	---
Povidone-iodine	22.3	15.4	7.1	5	0.0015 **
IPC	23.7	14.7	8.6	6.2	0.0021 **
SURFAXN	24.1	14.92	8.9	8.9	0.0020 **
<i>S. aureus</i>	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	---
Povidone-	26.5	15.9	9.2	3.5	0.0019

iodine					**
IPC	27.2	16.5	9.7	3.9	0.0018 **
SURFAXN	28.3	16.6	10.3	4.3	0.0018 **
<i>E. coli</i>	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	---
Povidone-iodine	26.5	14.82	8.1	3.6	0.0012 **
IPC	27.2	14.5	9.8	4.5	0.0010 **
SURFAXN	29.3	15.5	9.7	4.7	0.0012 **

Table 5 MIC of anti-biofilm disinfectants-CV after 16 hours

Weak	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	P-value
Povidone-iodine	54.6	33.1	15.6	10.3	0.0001 **
IPC	56.1	34.2	16.1	11.8	0.0001 **
SURFAXN	56.2	33.41	16.6	12.5	0.0001 **
Moderate	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	---
Povidone-iodine	44.375	28.7	13.5	10.5	0.0006 **
IPC	44.5	29.6	14.3	11.5	0.0005 **
SURFAXN	45.625	30.8	15.3	12.7	0.0006 **
Strong	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	---
Povidone-iodine	41.2	28.1	12.5	9.8	0.0017 **
IPC	42.4	29.2	13.1	10.5	0.0020 **
SURFAXN	41.5	30.7	12.5	11.1	0.0018 **
<i>S. aureus</i>	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	---
Povidone-iodine	43.3	26.9	13.5	10.5	0.0009 **
IPC	44.5	29.8	15.3	11.5	0.0008 **
SURFAXN	45.625	30.8	15.375	12.7	0.0009 **
<i>E. coli</i>	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	---
Povidone-iodine	45.3	27.75	13.5	11.5	0.0012 **
IPC	46.5	28.8	14.3	11.5	0.0009 **
SURFAXN	46.8	27.8	14.9	10.6	0.0009 **

Table 6 MIC of anti-biofilm disinfectants-CV after 24 hours

Weak	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	P-value
Povidone-iodine	62.7	41.3	20.1	13.3	0.0001 **
IPC	64.5	44	22	14.4	0.0001 **
SURFAXN	66.7	44.7	22.8	14.6	0.0001 **
Moderate	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	---
Povidone-iodine	51	38.8625	15.75	11.2	0.0001 **
IPC	51.625	39.625	16.25	12.3	0.0001 **
SURFAXN	55.375	39.25	17.375	14.2	0.0001 **
Strong	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	---
Povidone-iodine	48.1	37.4	14.2	9.6	0.0003 **
IPC	49.7	36.71428571	15.4	10.8	0.0001 **
SURFAXN	50.5	36.8	14.2	10.5	0.0001 **
<i>S. aureus</i>	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	---
Povidone-iodine	52	38.8	15.85	10.9	0.0001 **
IPC	51.6	39.6	16.45	12.2	0.0001 **
SURFAXN	54.3	39.2	17.3	11.2	0.0001 **
<i>E. coli</i>	2 MIC (mg/ml)	MIC (mg/ml)	1/2 MIC (mg/ml)	1/4 MIC (mg/ml)	---
Povidone-iodine	52.8	37.8	15.7	12.4	0.0001 **
IPC	50.6	38.6	16.2	13.2	0.0001 **
SURFAXN	52.3	39.2	17.3	13.5	0.0001 **

Using a povidone-iodine concentration of 0.2%, an antibacterial experiment demonstrated that IPC and surfaxn effectively stifled bacterial growth. All of the studied microorganisms have a minimum inhibitory concentration (MIC) of 10%, 5%, and 20%, respectively, for each chemical. This data suggests that the chemicals have the potential to reduce bacterial proliferation. Viability assay results utilizing CV shown that all drugs are effective after 2 hours of treatment and can continue their impact for up to 24 hours. According to the findings, these chemicals are effective against both Gram-negative and Gram-positive bacteria.

Infections induced by biofilms are a lot more worrying than those caused by planktonic cells. Biofilm-associated infections are notoriously difficult to treat because they necessitate larger

medication doses [10]

In addition to killing bacteria in 24 and 72 hours pre-formed biofilms, the povidone-iodine, IPC, and surfaxn complex showed no significant difference, increasing the likelihood that it is an effective antimicrobial agent. Not much is known about the process by which chemicals like povidone-iodine, indole-3-propionic acid (IPC), and surfaxn prevent biofilm formation[11], [12]

As more and more antimicrobials fail in the milk and dairy industries, causing economic loss and food safety difficulties, the proliferation of MDR *S. aureus* with high virulence has become a serious worry. Previous research has found that *S. aureus* isolated from milk is resistant to a wide variety of medicines. This includes erythromycin, ciprofloxacin, clindamycin, gentamicin, penicillin, and streptomycin. Furthermore, the majority of these isolates are multidrug-resistant (MDR) [13].

One way that *P. aeruginosa* stays alive in the face of antimicrobial agents and other stresses is by forming bacterial biofilms, which prevent the antimicrobials from penetrating the biofilm and killing the bacteria within it[14].

Multiple-drug resistance (MDR) and biofilm formation by *S. spp.* isolated from milk and dairy sectors have been linked in numerous studies. The MTT assay was used to determine the efficacy of chemicals in killing bacteria present in biofilms. To kill *P. aeruginosa* in biofilms, researchers tried three different compounds: povidone-iodine, isopropyl alcohol (IPA), and surfaxn [15], [16].

This setting was predicted to have minimal anti-biofilm activity. However, most of the evaluated formulations were only effective against Gram-negative germs, and only in the first 6 hours. After then, there was noticeable recovery linked to the original biofilm's leftover bulk, suggesting that the bacteria might be able to reform a new, more organized colony. Therefore, the entire colony needs to be eradicated in vivo, which requires debridement as well as repeated antiseptic treatments[17].

Bacterial Resistance Profile for Multi Antibiotics Agents

Nosocomial infections, which are acquired in hospitals, are notoriously difficult to treat and are often associated with *P. aeruginosa*. This is because this creature naturally resists pharmacological intervention. Moreover provided with the capacity to acquire additional resistance mechanisms to several types of antimicrobial drugs via horizontal gene transfer[18].

There was a wide range of antibiotic resistance among the bacterial isolates tested in this study, with meropenem showing the highest level of resistance (58 isolates, 98.3%), followed by imipenem (52 isolates, 88.1%), tobramycin and ofloxacin (51 isolates, 86.4%), gentamycin (44 isolates, 74.6%), ciprofloxacin (42 isolates, 71.2%), amikacin (35 isolate

Isolated *Acinetobacter spp.* and *P. aeruginosa* were tested for antibiotic sensitivity. Overall, 85.3% of the isolates were resistant to ceftazidime, and 73.50% were resistant to piperacillin-tazobactam, whereas only 11.88% were resistant to amikacin. Resistance to cefotaxime (93.8%), piperacillin-tazobactam (87.5%), and ceftazidime (87.5%) was more common among *Acinetobacter spp.*, while resistance to amikacin (12.5%) and ciprofloxacin (87.5%) was the lowest (18.8%). It was also found that *P. aeruginosa* isolates were highly resistant to ceftazidime (83.3%) and aztreonam (77.8%). However, 88.9% of *P. aeruginosa* isolates were killed by amikacin and 83.3% by meropenem [19].

Isolated bacteria were effectively treated with imipenem. Resistance to several medications was found in 26.5 percent of *P. aeruginosa* isolates [20].

Conclusions:

1. The results of this current study showed that the rate of isolation of *P. aeruginosa* bacteria isolated from burns and wounds equal to 59%.
2. It was found that all *P. aeruginosa* isolates isolated from burns, wounds, had varying resistance

to the antibiotics used in the study, the highest resistance was to antibacterial meropenem 58 isolates, 98.3% and the lowest resistance was to antimicrobial ampicillin 16 isolates, 27.1%.

3. The results of the study showed that povidone iodine and IPC231 have good efficacy against *P. aeruginosa* isolated from burns and wounds in terms of its effect at low concentrations in contrast with surfaxn.

References

1. Kim, M., Weigand, M. R., Oh, S., Hatt, J. K., Krishnan, R., Tezel, U. & Konstantinidis, K. T. 2018. Widely used benzalkonium chloride disinfectants can promote antibiotic resistance. *Applied and Environmental Microbiology*, 84(17): e01201-18.
2. Bragg, R. R., Meyburgh, C. M., Lee, J. Y. & Coetzee, M. 2018. Potential treatment options in a post-antibiotic era. *Infectious Diseases and Nanomedicine III*: 51-61. Springer, New Delhi.
3. Salter, S. J. 2015. Keeping an eye on *P. aeruginosa*. *Nature Reviews Microbiology*, 13(2): 69-69.
4. Klockgether, J. and Tümmler, B. 2017. Recent advances in understanding *Pseudomonas aeruginosa* as a pathogen. *F1000Research*, 6.
5. Bassetti, M., Vena, A., Croxatto, A., Righi, E. & Guery, B. 2018. How to manage *Pseudomonas aeruginosa* infections. *Drugs in context*, 7.
6. Voidazan, S., Albu, S., Toth, R., Grigorescu, B., Rachita, A., & Moldovan, I. (2020). Healthcare associated infections—a new pathology in medical practice?. *International journal of environmental research and public health*, 17(3), 760.
7. Clutterbuck, A. L., Woods, E. J., Knottenbelt, D. C., Clegg, P. D., Cochrane, C. A. & Percival, S. L. 2007. Biofilms and their relevance to veterinary medicine. *Veterinary Microbiology*, 121(1-2): 1-17.
8. Anonymus. 2016. Clinical and Laboratory Standards Institute (CLSI). Performance Standards for Antimicrobial Susceptibility Testing; Twenty- Six Informational Supplement. CLSI Document M 100- S26. Wayne, PA: Clinical and Laboratory Standards Institute.
9. Shafiee, M., Lotfi, F. H. & Saleh, H. 2014. Supply chain performance evaluation with data envelopment analysis and balanced scorecard approach. *Applied Mathematical Modelling*, 38(21-22): 5092-5112.
10. Liu, H., Zhao, Y., Zhao, D., Gong, T., Wu, Y., Han, H. & Qu, D. 2015. Antibacterial and anti-biofilm activities of thiazolidione derivatives against clinical staphylococcus strains. *Emerging Microbes & Infections*, 4(1): 1-6.
11. Ghosh, C., Manjunath, G. B., Konai, M. M., Uppu, D. S., Hoque, J., Paramanandham, K. & Haldar, J. 2015. Aryl-alkyl-lysines: agents that kill planktonic cells, persister cells, biofilms of MRSA and protect mice from skin-infection. *PLoS One*, 10(12): e0144094.
12. Olsen, I. 2015. Biofilm-specific antibiotic tolerance and resistance. *European Journal of Clinical Microbiology & Infectious Diseases*, 34(5): 877-886.
13. Founou, L. L., Founou, R. C. & Essack, S. Y. 2016. Antibiotic resistance in the food chain: a developing country-perspective. *Frontiers in Microbiology*, 7: 1881.
14. Rabin, N., Zheng, Y., Opoku-Temeng, C., Du, Y., Bonsu, E. & Sintim, H. O. 2015. Biofilm formation mechanisms and targets for developing antibiofilm agents. *Future Medicinal Chemistry*, 7(4): 493-512.
15. Wang, W., Lin, X., Jiang, T., Peng, Z., Xu, J., Yi, L. & Baloch, Z. 2018. Prevalence and characterization of *Staphylococcus aureus* cultured from raw milk taken from dairy cows with mastitis in Beijing, China. *Frontiers in Microbiology*, 9: 1123.

16. Meroni, G., Soares Filipe, J. F., Drago, L. & Martino, P. A. 2019. Investigation on antibiotic-resistance, biofilm formation and virulence factors in multidrug resistant and non multi drug resistant *Staphylococcus pseudintermedius*. *Microorganisms*, 7(12): 702.
17. Wilkins, R. G. and Unverdorben, M. 2013. Wound cleaning and wound healing: a concise review. *Advances in Skin & Wound Care*, 26(4): 160-163.
18. Strateva, T. and Yordanov, D. 2009. *Pseudomonas aeruginosa* a phenomenon of bacterial resistance. *Journal of Medical Microbiology*, 58(9): 1133-1148.
19. Mekonnen, H., Seid, A., Molla Fenta, G. & Gebrecherkos, T. 2021. Antimicrobial resistance profiles and associated factors of *Acinetobacter* and *Pseudomonas aeruginosa* nosocomial infection among patients admitted at Dessie comprehensive specialized Hospital, North-East Ethiopia. A cross-sectional study. *Plos One*, 16(11): e0257272.
20. Samad, A., Ahmed, T., Rahim, A., Khalil, A. & Ali, I. 2017. Antimicrobial susceptibility patterns of clinical isolates of *Pseudomonas aeruginosa* isolated from patients of respiratory tract infections in a Tertiary Care Hospital, Peshawar. *Pakistan Journal of Medical Sciences*, 33(3): 670.