©NIJSES
Publications

Open Access
ONLINE ISSN: 2992-5819
PRINT ISSN: 2992-6149

NEWPORT INTERNATIONAL JOURNAL OF SCIENTIFIC AND EXPERIMENTAL SCIENCES (NIJSES)

Volume 4 Issue 1

https://doi.org/10.59298/NIJSES/2023/10.7.1000

Efficacy Assessment of Some Commonly used Disinfectants against Multidrug Resistant Bacterial Isolates from a Tertiary Healthcare Facility in Benin City, Nigeria

Samson O. Onemu[1], Oluyemisi Odeyemi[2], Matthew Eturhobore Adu[3] and *Emmanuel Ifeanyi Obeagu[4]

[1]Samson O. Onemu, Department of Medical Laboratory Science, Achievers University, Owo, Nigeria
[2]Oluyemisi Odeyemi, Department of Medical Laboratory Science, Achievers University, Owo, Nigeria.
[3]Michael O. Adu, Department of Chemical Sciences, University of Delta, Agbor, Nigeria,
[4]Department of Medical Laboratory Science, Kampala International University, Uganda.
***Corresponding author: Emmanuel Ifeanyi Obeagu, Department of Medical Laboratory Science, Kampala International University, Uganda, emmanuelobeagu@yahoo.com, obeagu.emmanue@kiu.ac.ug, 0000-0002-4538-0161**

ABSTRACT

The ever-rising numbers of multidrug resistant, MDR bacteria in healthcare facilities globally has correspondingly led to a rapid escalation in the use of disinfectants, antiseptics or biocides which reached unprecedented levels during the COVID-19 pandemic. This has also elicited grave apprehension about the development of resistance to disinfectants. This study was conducted to evaluate the effectiveness of some regularly used disinfectants in the study population. Five brands of disinfectants were obtained and diluted in sterile 300 ppm calcium carbonate-nutrient broth to their respective minimum inhibitory concentrations, MICs to which were exposed broth cultures of MDR organisms for 2 min before sampling onto nutrient agar plates in replicates. Further samples were taken

from the mixture at 4, 6, 8, … 30 min intervals. One set of plates were incubated at 37°C and the other at RT for 48 h and 96 h respectively and examined for growth. Sodium hypochlorite, phenol and glutaraldehyde - all proved to be effective with the fastest time kill kinetics at 2.5 min. The longest time kill kinetics were demonstrated by cetrimide and cresol against *Pseudomonas aeruginosa* at 16 min and 13 min respectively, thus affirming Pseudomonas aeruginosa as the most resistant organism. This study infers of a reassurance in the continued effectiveness of the disinfectants under review in the phase of the ever-soaring tide of MDR organisms in healthcare settings.
Keywords: Disinfectants, usage, effectiveness, MDR-organisms.

INTRODUCTION

The emergence of methicillin resistant *Staphylococcus aureus*, MRSA, culminated in the incremental use of disinfectants in hospitals and public places since the 1960s [1-7]. An astronomical escalation in the application of disinfectants, antiseptic or biocides worldwide followed the SARS-CoV-2 (or COVID-19) pandemic [8-10] and the resultant existence of an intense atmosphere of health insecurity [11]. Disinfectants are regarded as having a critical role in the control and blotting the chain of spread of infectious agents in the hospital and community. [12]. An ever-rising frequency of multidrug resistant, MDR pathogenic bacterial species that are resistant to most of the available antibiotics has aroused tremendous interest and concern worldwide on the ability of the healthcare systems of communities to cope with the onslaught of an eventual MDR organism-induced pandemic with no readily available treatment option as was the case during SARS-COVID-19 pandemic [13-16]. Multidrug resistant bacterial species are usually organisms that have become resistant to a wide spectrum of antibiotics [17]. Resistant bacterial phenotypes are known to be demonstrable in all important groups of bacteria that initiate human disease [18] which also heightens the risks of contamination to the environment [19]. Disinfectants, antiseptics or biocides have extensively been employed in hospital care, homes and in aerial decontamination of public places that possess the potential for transmission of infectious agents [20-21]. The specific dangers of contaminated environment lie on the potential for transference of infectious materials from one patient to another in a hospital environment or within a community and the attendant consequences of hospital acquired infections, HAIs [22]. Current observations indicate that HAIs cases have rising steeply in hospitals worldwide [23-24], and they are key reasons for longer hospitalization periods, higher health care costs and increased disease burden and elevated death tolls [25]. Recent literatures suggest that MDR organisms have developed resistance genes to disinfectants [26] or the increasing use of disinfectants may also confer cross-resistance to antibiotics). The activity of many disinfectants on bacteria is targeted at the proteins in the cell-wall that leads to the disruption of the cell's permeability [27] whereas, antibiotics resistance focuses on target site alteration, development of alternative enzymes, drug destroying enzymes and use outflow pumps within a bacterium to export harmful substances out of the cell [18]. This study was planned to investigate whether decontamination can be safely achieved in an environment defiled by multidrug resistant bacteria using commonly available disinfectants in the study population.

MATERIALS AND METHODS

Twenty MDR isolates each of *Escherichia coli*, *Klebsiella* species, *Proteus* species, *Pseudomonas aeruginosa*, *Citrobacter* species, *Alcaligenes faecalis*, *Staphylococcus aureus* and *Enterococcus faecalis* were collected from a tertiary healthcare facility in Benin City, Nigeria, and five different types of commonly used disinfectant brands namely: Cetavlon (cetrimide), Hypo (sodium hypochlorite), Dettol (phenol), Wavicide (glutaraldehyde) and Izal (cresol) were procured and diluted to their respective minimum inhibitory concentrations, MICs following the manufactures instructions in sterile 300 ppm calcium carbonate ($CaCO_3$) nutrient broth to simulate a moderately dirty environment. Each diluted disinfectant was dispensed in 3.0 mL volumes in sterile universal bottles, to a 3.0 mL diluted disinfectant was added 1.0 mL of an 18 h broth culture of MDR isolate, mixed and a timer was set. At the lapse of 2.0 minutes 0.1 mL of the mixture was removed and dispensed onto a dried nutrient agar plate in replicate and spread with a sterile loop. This procedure was repeated at 4, 6, 8, 10 …30 minutes. The 18 h broth culture of each MDR isolate was diluted 1:100 in sterile 300 ppm $CaCo_3$ nutrient broth and 0.1 mL was also inoculated in replicates onto another set of dried nutrient agar plates as positive control. One set of plates were incubated at 37°C for 72 h and the other set at room RT for 96 h. The tests plates were examined for growth in comparison with the positive control test plates.

RESULTS

The data from Table 1 illustrates the mean kinetic kill times of various disinfectants. Sodium hypochlorite, glutaraldehyde, and phenol emerged as the fastest in terms of kill rates, taking an average of 2.5 minutes to eliminate all MDR isolates, encompassing Pseudomonas aeruginosa, Staphylococcus aureus, and Enterococcus faecalis. Following closely, cresol exhibited a notable activity, achieving a maximum kill time of 13.0 minutes against Pseudomonas aeruginosa and Alcaligenes faecalis. Cetrimide demonstrated variable kill times, ranging between 12.5 to 16.0 minutes, showcasing the longest kill time specifically for Pseudomonas aeruginosa

Open Access
ONLINE ISSN: 2992-5819
PRINT ISSN: 2992-6149

Table 1: Mean time kill kinetic of disinfectants (minutes)

MDR Organism	Disinfectants				
	Sodium hypochlorite (1:20)	etrimide (1:50)	Glutaraldehyde (1:100)	Phenol (1:50)	Cresol (27:973)
Escherichia coli	2.4	12.5	2.5	2.5	11.5
Klebsiella species	2.5	12.5	2.5	2.5	11.5
Proteus species	2.5	12.5	2.5	2.5	11.5
Pseudonons aeruginosa	2.5	16	2.5	2.5	13.0
Citrobacter species	2.5	12.5	2.5	2.5	10.0
Alcaligenes faecalis	2.5	12.5	2.5	2.5	10.0
Staphylococcus aureus	2.5	12.5	2.5	2.5	11.5
Enterococcus faecalis	2.5	125	2.5	2.5	11.5

DISCUSSION

The different disinfectants evaluated proved effective in eliminating MDR organisms. The fastest time kill kinetic were demonstrated with sodium hypochlorite (domestic bleach), brands of phenol (Izal) and glutaraldehyde (Wavicide), each with a mean time kill kinetic of 2.5 minutes. A disinfectant is required to accomplish the elimination of pathogens especially those that initiate disease within 10 minutes which has been achieved with this disinfectant [28]. Studies have indicated that sodium hypochlorite is one of the most rapid acting disinfectants [29]. The highest time kill kinetic of 16.0 minutes was observed with cetrimide against *Pseudomonas aeruginosa.* This disinfectant is recognized to permit the growth of *Pseudomonas aeruginosa* in-use dilutions [30-31] and as a precautionary measure, its application should therefore, be discouraged in areas where contamination with *Pseudomonas aeruginosa* is anticipated. This is partly due to the notoriety of this microorganisms to be resistant to many chemical agents and antibiotics and a major organism associated with hospital acquired infections (HAIs). This organism has as a consequence been designated as an organism of prime relevance by the World Health Organization [32-33]. The findings from this investigation are further indication that there is no direct proof of disinfectants resistance as seen in antibiotics as other studies have inferred [28]. The non-selectivity of the mechanism of action of disinfectants against bacteria appear to play a significant role in toxicity of disinfectants by interference with bacterial cell membrane proteins thereby disrupting permeability of bacteria cell-walls [34], whereas antibiotics are selectively toxic to specific biochemistries in the microorganism. It is clearer therefore, to visualize that bacteria acquisition of resistance to antibiotics should be tremendously easier to navigate and faster to accomplish in comparison to disinfectants [18]. Failure of disinfectants have been largely traceable to incorrect dilutions which are common-place and inadequate or misapplication that have been highlighted [35].

CONCLUSION

The disinfectants evaluated in this study remained effective in eliminating MDR organisms, and there was no direct evidence to suggest that the ever-growing incidence of MDR organisms will also translate to corresponding resistance to disinfectants. It is reemphasized however, that disinfectants should be used following the manufacturers' guidelines.

REFERENCES

1. Maillard, J. Y. and Pascoe, M. (2023). Disinfectants and antiseptics: Mechanisms of action and resistance. *Nat. Rev. Mcirobol.* doi.org/10.1038/s41579-023-00958-3.
2. Chukwueze, C. M., Okpala, O. V., & Obeagu, E. I. (2022). A systematic review on Methicillin resistant Staphylococcus aureus in patients with surgical wounds. *Int. J. Adv. Multidiscip. Res, 9*(9), 25-36.

Open Access
ONLINE ISSN: 2992-5819
PRINT ISSN: 2992-6149

3. Chukwueze, C. M., Okpala, O. V., & Obeagu, E. I. (2022). Prevalence of Methicillin resistant Staphylococcus aureus in patients with surgical wounds attending Esuth, Parklane. *Int. J. Adv. Multidiscip. Res*, *9*(9), 1-12.
4. Onemu, S. O., Ademulegun, F., Onemu-Metitiri, M. O., Obeagu, E. I., & Hassan, A. O. (2023). The Contribution of Curable Plasmid-Mediated Resistance in Isolates of Staphylococcus aureus at the University of Benin Teaching Hospital, Benin City, Nigeria. *Asian Journal of Dental and Health Sciences*, *3*(3), 30-32.
5. Chukwueze, C. M., Udeani, T. K., Obeagu, E. I., & Asogwa, N. (2022). ANTIBIOTIC SUSCEPTIBILITY PATTERN OF METHICILLIN RESISTANT STAPHYLOCOCCUS AUREUS IN HOSPITALIZED WOUND PATIENTS IN SELECTED TERTIARY HOSPITALS IN ENUGU METROPOLIS.
6. Chukwueze, C. M., Obu, J. O., & Obeagu, E. I. (2023). Isolation and identification of bacteria from surgical wound attention at Enugu State University Teaching Hospital, Enugu. *Int. J. Curr. Res. Chem. Pharm. Sci*, *10*(8), 35-43.
7. Onemu, S., Ige, R., Onemu-Metitiri, M., Obeagu, E. The prevalence of asymptomatic bacteriuria in pregnant women in Akure, Ondo State, Nigeria. *DYSONA – Life Science*, 2024;5(1): 1-8. doi: 10.30493/dls.2023.151009
8. Dewey, H. M., Jones, J. M. Keating, M. R. and Uprety I. B. (2021). Increased use of disinfectants during the COVID-19 pandemic and its potential impacts on health and safely *ACS Chem. Health Sci.* **29**(1): 27-38.
9. Ghofoor, D, Khan, Z., Khan, A. Ualiyeva, D. and Zaman, N. (2021). Excessive use of disinfectants against COVID-19 posing a potential threat to living things. *Curr. Res. Toxicol.* **2**:159-168.
10. Marteinson., S.C. Taylor, J.J., Green, A, Winegardner A. K., Rytwinski, T, Reid, J. L., Dubertz C, Lablanc, J., Gaius, M. D. and Cooke, S. J. (2022). Increased use of disinfectants during the COVID-19 pandemic: Identification of antimicrobial chemicals and consideration for aquatic environmental contamination. *Environ. Rev.* (2022): doi.org/10.//39/er-2023-0035.
11. Chen, Z., Guo, J., Jiang, Y. and Shao, Y. (2021). High concentration and high dose of disinfectants and antibiotics used during COVID-19 pandemic threaten human health. *Environ. Sci. Eur.* **33**(11): doi.org/10.1186/s12302-021-00456-4.
12. Montagna, M. T., Triggiano, F., Barbuti, G., Bartolomeo, N., De-Giglio, S., Diela, G., Lopuzzo, M., Rutigliano, S., Serio G..and Caggiano, G. (2019). Study on *In-Vitro* activity of five disinfectants against nosocomial bacteria. *Int. J. Envrion. Res. Public Health* **16**(11): 1895.dio3390/ijerph.1611185.
13. Bassetti, M., Pecori,. D. and Peghin M (2016). The mangement of multidrug resistant enterobacteriaceae: *Curr. Opin. Infect. Dis.* **29**(6): 583-594 doi.1097QCD.0000000000314.
14. Perez-Etayo, L., Gonzalez, D., Leiva, J. and Vitas, A. I. (2020). Multidrug resistant bacteria isolated from different aquatic environments in the North of Spain and South France. *Microorganisms* **8**(9): 1425 doi.10.3390/microorganism. 1425.
15. Jimenez, M. C., Kowalski, L., Souto, R. B., Alves, I. A., Vianna, M. D. M., and Aragon, D.A. (2022). New drug against multidrug resistant bacteria: A systematic review of patients. *Future Microbiol.* **17**(17): doi.org/10.2217/tmb.2022.0104.
16. Najeem, S. Eick, D., Boettcher, J., Aigner, A., Aboutara M., Fenner, I., Reinshagen, K. and Koenigs I. (2022). High prevalence of multidrug resistant Gram-negative bacteria carriage in children screened prospectively for multidrug resistant organisms at admission to a pediatric hospital, Hamburg, Germany. *Euro. Surveill.* **27**(15): doi.org/10.2807/1560-7917.ES.2022.27.15.202001567.
17. Kumar, V. A and Khan, D. (2015). Defining Multidrug resistance in Gram-negative bacilli. *Indian J. Med. Res.* **141**(4): 491-492 doi.10:410369715916.159318.
18. Weber, D. J., Rutal, W A., and Sickbert-E. S., (2019). Use of germicide in health care settings. Is there a relationship between germicide use and antimicrobial resistance: A concise review. *Am. J. Infect. Control.* **475**: A106-A109 doi.1016/ajic.2019.03.023.
19. Fard, N. J., Jorfi, S., Fard, M. P., Zadeh M. A. (2021). The effect of the use of disinfectants during COVID-19 pandemic on the bacterial contamination of dental unit waterlines. *Environ. Health Engineering Management J.* **9**(3): 255-260.
20. Perry-Dow, K. A., De-Man, T .J. B., Halpin, A. L., Shans. A., Rose. L. J. and Noble-Wang. J. (2022). The effect of disinfectants on the microbial community in environmental health care surfaces using next generation sequencing. *Am. J. Infect. Control* **50**(1): 54-60.
21. Chacon-Jimenez, L and Rojas-Jimenez, K. (2020). Resistance to disinfectant and their relationship to antibiotic resistance. *Acta Med. Geriatric* **62**(1): 1-12. 2580 0001-6002.

Open Access
ONLINE ISSN: 2992-5819
PRINT ISSN: 2992-6149

22. Tong, C., Hu, H., Chem, G., Liz, Li, Z. and Zhang, J. (2021). Disinfectant resistance in bacteria: Mechanisms, spread and resolution strategies. *Environ. Res.* **195**:110 897 doi.orgl10.1016/j.envres 2021.
23. Alajlan, A.A., Mukhtar, L. E., Almussallam, A. S., Alnuqaydan A. M., Abakiri, N. S., Al-Mutari, T. F., Bin-Shahail K. M and Aldawsari, F. S and Alajel, S. M. (2021). Assessment of disinfectants efficacy in reducing microbial growth PLoS ONE. **17**(6): e0269850.
24. Fernandez- Martinez, N.F., Carcel-Fernadez, S., Fuento-Martos, R., Ruuz-Montero R., Guzman-Herrado B.R., Leon-Lopez, R., Gomez, F. J., Guzman-Puche, J and Salcedo-Lea, I. (2022). Risk factor for multidrug resistant Gram-negative bacteria carriage upon admission to intensive care unit. *Int. J. environ. Res. Public Health.* **19**(3): 1039 dio.org/10/ijerph.19031039.
25. Schumacher, M., Allignol, A., Beyersmann, J., Binder, N. and Wolkewitz, M. (2013). Hospital acquired infection: Appropriate statistical treatment is urgently needed. *Inter J. Epiemiol.* **42**(5): 1502-1508 doi.org/10. 1093/ije/dyt/111.
26. Schinoda, N., Mitarai, S. Suzuk,i E and Watanabe, M. (2016). Disinfectant susceptibility of multidrug resistant *Mycobacterium tuberculosis* isolated in Japan. *Antimicrobial Resistance and Control* **5**(3): doi.org/10.1186/s13756-016-0102-y.
27. Denyer S. P. and Stewart, G. S. A. B. (1998). Mechanism of action of disinfectants. *Int. Biodeteroration Biodegradatiion.* **41**(3-4): 261-268.
28. Rutala, W. A. and Weber, D. J. (2015). *Manell, Doglas and Bennett's Principles and Practice of Infectious Disease.* 2015:3294-3309.e4. doi.1016/B978-1-4557-48013.003015.
29. Agbo, E. C., Achi, O. K., Nwachukwu, E., Obeta M. U., Obiora, E. O., Maduka, K. M., Oraekeyi N. P and Lote-Nwaru, I. E. (2020). Time kill kinetics study of commonly used disinfectants against biofilm forming pseudomonas aerugmosa in Federal Medical Centre, Umuahia, Nigeria. *Am J. Biomed. Sci. Res.* **7**(3): doi.10.34297/AJBSR.2020.07-001155.
30. Boyce J. M., and Hawill, N. L (2022). In-use contamination of a hospital grade disinfectant. *Am. J. Infect. Control.* **50** (12): 1246-1301.doi.10.1016/mjic 2022031008. 110897.
31. Lompo, H., Herees, A., Agbabli E., Kazenga, A., Peeters M., Tinto, H., Lagram, K., Sangare, L, Affolabi, D, and Jacobs, J. (2023). Growth of gram-negative in antiseptics, disinfectants and hand hygiene products in two tertiary hospitals: A cross sectional survey. *Pathogens* **12**(7): doice.3390/patogens 12070917.
32. Langendonk, R. K, Neill, D. R. and Forth Ergill J. L., (2021). Pseudomonas aeruginosa, Pathogenesis, Vigilance, Antibiotic Tolerance and Resistance, stress responses and host-pathogen interactions. *Front. Cell. Infect. Microbial.* 11.2021 doi.org/10.3389/femib.2021.66759.
33. Bakht, M., Alizadeh, S. A., Rahimi, S., Anari, R.K., Rostamani, M., Javadi, A., Peymani, A., Marshi S. M A. and Nikkhahi F (2022). Phenotype and genetic determination of resistance to common disinfectants among biofilm producing pseudomonas aemgmosa stain from clinical specimens in Iran. *BMC- Microbial.* 22:124 doi.org/10.1186/s12866.022.02524-y.
34. Zhang K. W., Hu, H and Chem, G (2022). Mechanism of microbial disinfectant resistance. *Progress Biochem. Biophys.* **49**(1): 34-47.
35. Jones, I. A. and Joshi, L. T. (2021). Biocide use in the antimicrobial era: A review: *Molecules* **26**: 2276 doi.org/10.3590/molecules.26082276.

CITE AS: Samson O. Onemu, Oluyemisi Odeyemi, Matthew Eturhobore Adu and Emmanuel Ifeanyi Obeagu (2023). Efficacy Assessment of Some Commonly used Disinfectants against Multidrug Resistant Bacterial Isolates from a Tertiary Healthcare Facility in Benin City, Nigeria. NEWPORT INTERNATIONAL JOURNAL OF SCIENTIFIC AND EXPERIMENTAL SCIENCES (NIJSES) 4(1):48-52.
https://doi.org/10.59298/NIJSES/2023/10.7.1000