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The Effect of some Disinfectants and Antiseptics on Pseudomonas Bacteria Isolated from the Environment of Al-Kindi Hospital in Baghdad

Salwa Ghali Bdawi
QA. Mustansiriyah University
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Abstract

This study aims to evaluate the prevalence of Pseudomonas spp. in hospital environments and their resistance to commonly used disinfectants, given their significant role in healthcare-associated infections. A total of 132 swabs were collected from various surfaces at Al-Kindy Hospital between August 2024 and March 2025, yielding 46 Pseudomonas isolates out of 260 bacterial isolates. Susceptibility testing of these isolates against several commercial disinfectants revealed variable effectiveness, with notable resistance especially in isolates from medical devices and frequently touched surfaces. This resistance is attributed to the bacteria's ability to form biofilms, which enhance their tolerance to disinfectants. The study highlights the critical need to review disinfection protocols and ensure proper use and regular evaluation of disinfectant efficacy to control the spread of resistant organisms and safeguard patient safety in healthcare settings.

Keywords: Pseudomonas, Disinfectants, Zone of inhibition, Isolates, Resistance.

* Introduction

Disinfectants play an important role in controlling the spread of infectious diseases caused by bacteria and spore-forming organisms, as both bacteria and spores can persist on surfaces and in the environment for long periods, posing a significant risk to public health. The rate of hospital-acquired infections has increased significantly worldwide (Caitlinn B. Linebck et al., 2018).

The emergence of resistance to disinfectants has become a life-threatening issue due to the reduced effectiveness of these agents. According to the Centers for Disease Control and Prevention (CDC), approximately two million people may become infected annually, with around 950 deaths resulting from such infections (Chaoyu, 2021).

The morbidity and mortality associated with hospital-acquired infections are extremely high. In the United States alone, more than two million infections occur among infants, with 50–60% of these infections being caused by resistance to antibiotics and disinfectants.

It is also estimated that between 9,600 and 20,000 patients die annually in the United States due to bloodstream infections associated with hospital-acquired infections (Al-Ratib et al., 2021).

The global spread of antibiotic-resistant bacteria indicates a growing prevalence of these pathogens. Antibiotic resistance has emerged as a new disease worldwide due to its rapid transmission. This has created an urgent need for effective sterilization products, as disinfectants can effectively control infectious diseases by inhibiting or destroying the growth of pathogens, including bacteria and viruses (Chaoyu, 2021).

Among the most frequently occurring Pseudomonas species in hospital-acquired infections is Pseudomonas aeruginosa, which has become one of the most common pathogens, especially in immunocompromised patients. It is responsible for 10–20% of hospital-acquired infections in intensive care units (AL-Ratib A., et al., 2021).

Pseudomonas is a motile, Gram-negative bacterium and is considered one of the major opportunistic pathogens responsible for various clinical infections in both animals and humans (Machado et al., 2013).

Pseudomonas adapts to antibiotics and disinfectants by forming biofilms on contaminated surfaces. These microbial biofilms enhance bacterial resistance to antimicrobial agents such as antibiotics and disinfectants (Drenkard, 2003).

The use of disinfectants in hospitals is considered an essential tool for combating the spread of infectious diseases when used properly. According to guidelines, appropriate disinfection protocols in hospitals and healthcare settings are crucial to reducing the risk of infections, especially given the increasing spread of these organisms.

In recent years, hospital-acquired infections have contributed significantly to increased morbidity and mortality worldwide. In the United States, more than 1.7 million infections occur annually, with approximately 100,000 deaths, largely due to microbial infections in healthcare settings. According to a report published by the European Centre for Disease Prevention and Control (ECDC), there are 33,000 deaths annually in the European Union due to multidrug-resistant bacterial infections (Naif et al., 2023).

*** Methodology**

This study was conducted from August 2024 to March 2025. A total of 132 swabs were collected from various sites in Al-Kindi Hospital using sterile swabs, including door handles, stair railings, medical equipment and devices (incubators, scales, blood pressure monitors), laboratory tables, patient beds, and the hands of healthcare workers. From these swabs, 260 bacterial isolates were obtained, of which 46 were identified as belonging to Pseudomonas species.

The swabs were placed in tubes containing sterile saline solution and immediately transported to the laboratory. Each swab was directly cultured on blood agar and MacConkey agar media. The plates were then incubated at 37°C for 18 to 24 hours. After incubation, the plates were examined for typical Pseudomonas colony characteristics and pigment production. Additional tests, including the oxidase test and the API 20 test, were also performed.

A bacterial suspension was prepared and evenly spread onto Mueller-Hinton agar using a sterile swab to ensure full surface coverage. The plates were left to dry for 10 minutes.

The tested disinfectants—sodium hypochlorite, ethanol, Decosept, Cidex OPA, hydrogen peroxide, Hibitane, iodine solution, and Dettol—were diluted according to standard concentrations. Wells were created in the agar using a sterile tool, and 100 µL of each disinfectant was added into each well using a sterile micropipette, following the well diffusion method. Plates were left at room temperature for one hour to allow for diffusion, then incubated at 37°C.

After the incubation period, the diameters of the inhibition zones around each well were measured at intervals of 18, 24, and 36 hours. The results were recorded to compare the effectiveness of the different disinfectants.

Table 1: Disinfectants Used in the Study

Trade Name	Chemical Formula & Name	Concentration	Ingredients / Additives (if any)	Country of Manufacture
Ethanol alcohol	Ethyl alcohol CH_3CH_2OH	70%	Diluted with water	China
SODIUM HYPOCHLORITE	Sodium hypochlorite $NaClO$	0.5 – 1%	Sodium salts of hypochlorous acid	Turkey
Dettol	C_8H_9ClO	3%	Chloroxylenol	Iraq
Hibitane	$C_{22}H_{30}Cl_2N_{10}$	2 – 4%	Chlorhexidine digluconate	Syria
Deco Sept	Active ingredient: Isopropanol	80%	Propane-1-ol, Propane-2-ol	Germany
Cidex OPA	$C_8H_6O_2$	2 – 4%	Orthophthalaldehyde	—
Hydrogen Peroxide	H_2O_2	3%	Chloroxylenol	Syria
Iodine Solution	$(C_6H_9NO)_n \cdot xI_2$	10%	Polyvinylpyrrolidone	Syria

* Discussion

Figure 1 shows the proportions of Pseudomonas bacterial species isolated from various locations in Al-Kindi Hospital. It is noticeable that the prevalence of these species in hospitals plays a significant role in medical microbiology due to their association with hospital-acquired infections.

Pseudomonas bacteria, especially Pseudomonas aeruginosa, are among the most common opportunistic pathogens in hospital environments. The isolation rate in this study was 66.39%, which was the highest among all types, particularly in the intensive care unit. This is due to their increasing resistance to antibiotics and disinfectants, facilitating their spread (Hossein Fazeli, et al., 2012).

The prevalence of the second species, Pseudomonas fluorescens, in the hospital environment was 23.74%. Isolating this species is very rare in clinical or industrial environments compared to P. aeruginosa, because it is primarily found in natural environments such as soil and cold water and is considered non-pathogenic (Baader A and Garre, 1987). Although less virulent than P. aeruginosa, it can cause severe (opportunistic) infections in humans.

The isolation rate of Pseudomonas putida was 9.87%. This lower number compared to other Pseudomonas species is attributed to its lower pathogenicity and ability to cause infection, being a rare cause of clinical infections and sensitive to most antibiotics (Yusuke, et al., 2011). P. putida lives in hospital environments and occasionally causes hospital infections in severely ill or immunocompromised patients, but isolating strains of P. putida is rare (Carpenter, 2008).

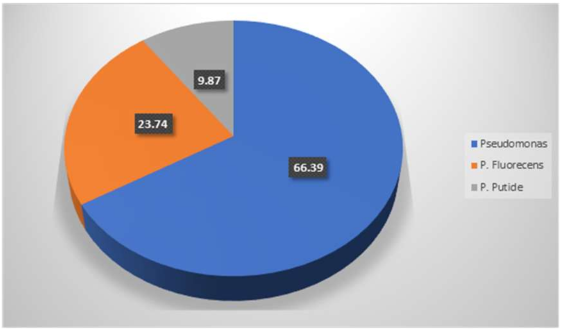


Figure 1: Proportion of Isolated Pseudomonas Species

Disinfectants and antiseptics can be sources of healthcare-associated infections, and Pseudomonas species are considered the most commonly isolated bacteria from contaminated disinfectants (William A. et al., 2008). This study tested the sensitivity of Pseudomonas species to several disinfectants, showing notable differences in inhibition zones, reflecting the effectiveness of those disinfectants and indicating the bacteria's resistance levels.

The sensitivity of Pseudomonas aeruginosa to disinfectants was tested after 18 hours of exposure. The highest efficacy was for (Cidex OPA) and (Sodium hypochlorite), with inhibition zones of 22 mm and 21 mm, respectively. This is because (Cidex OPA) is a highly effective disinfectant used for sterilizing medical instruments (CDC, 2008; RUTALA WA, et al., 2016). This matches findings from Al-Shater & Mahfouz (2022), who proved that Cidex OPA is the most efficient disinfectant against bacterial isolates at concentrations of 100%, 50%, and 25%. It disrupts bacterial proteins and enzymes leading to death. Sodium hypochlorite destroys bacterial cell components such as nucleic acids and proteins (RUTALA WA, et al., 2016).

Hydrogen peroxide showed good efficacy against P. aeruginosa with an inhibition zone of 18 mm, consistent with Cordoba LK et al. (2018), who demonstrated its effectiveness at certain concentrations against multiple bacteria including Pseudomonas species. Hibitane also had good efficacy with an 18 mm inhibition zone due to its action disrupting bacterial cell membranes causing leakage of cell contents (Asem M. Abdelshafy, 2024).

Iodine, Dettol, and Deco Sept showed moderate efficacy with inhibition zones of 15 mm, 16 mm, and 17 mm respectively. This may be due to partial bacterial resistance or insufficient concentrations (Hancock, R.E.W. & Speert, D.P., 2000).

Ethanol showed weak activity against P. aeruginosa with an inhibition zone of only 5 mm. This is attributed to the presence of an outer membrane in Gram-negative bacteria despite ethanol's high efficacy against many microorganisms.

For Pseudomonas fluorescens, disinfectants Cidex OPA, Deco Sept, Sodium hypochlorite, Hydrogen peroxide, and Hibitane were highly effective, showing inhibition zones of 24 mm, 21 mm, 21 mm, 20 mm, and 20 mm, respectively. Dettol and Ethanol showed weak effects with inhibition zones of 10 mm and 14 mm respectively. This aligns with AL-Ratib A. et al. (2021), who reported weak Dettol efficacy (10 mm) and diminishing Ethanol effectiveness, explained by Dettol's limited activity against Gram-negative bacteria due to their resistant cell structure (McDonnell, G., 1999).

Iodine was very weak, almost ineffective (4 mm), despite being a broad-spectrum disinfectant. This resistance is due to bacterial defense mechanisms that reduce the impact of oxidizing agents like iodine (Hou, A.-M, et al., 2024).

Pseudomonas putida showed high sensitivity to Cidex OPA, Deco Sept, Sodium hypochlorite, Hydrogen peroxide, and Hibitane with inhibition zones of 28 mm, 21 mm, 21 mm, 21 mm, and 21 mm, respectively. Dettol and Ethanol showed weak effects (14 mm and 10 mm). This somewhat agrees with previous studies confirming resistance to Ethanol at all concentrations (Al-Talib H., 2019). Iodine showed no effect after 18 hours, possibly due to insufficient contact time, highlighting the importance of exposure time in disinfectant efficacy (CDC, 2008).

Table 2: Effect of Disinfectants on Pseudomonas Species After 18 Hours

Pseudomonas	Ethanol alcohol	Sodium hypochlorite	Dettol	Deco sept	Cidex OPA	Peroxide hydrogen	Iodine solution	Hibitane
P. aeruginosa	5 mm	21 mm	16 mm	17 mm	22 mm	18 mm	15 mm	18 mm
P. fluorescens	12 mm	21 mm	8 mm	21 mm	24 mm	19 mm	4 mm	20 mm
P. putide	10 mm	21 mm	14 mm	20 mm	25 mm	20 mm	-	21 mm

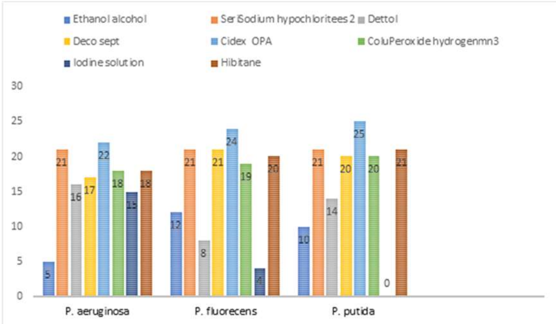


Figure 2: Effect of Disinfectants on Pseudomonas Species After 18 Hours

Efficacy generally increased with time, with Cidex OPA and Sodium hypochlorite showing the greatest effect on P. aeruginosa (25 mm and 22 mm inhibition zones). Hydrogen peroxide and Hibitane also showed good activity (19 mm each). Deco Sept, Dettol, and Iodine showed variable moderate to good effects (18 mm, 17 mm, and 16 mm).

For P. fluorescens, Cidex OPA, Sodium hypochlorite, Deco Sept, Hibitane, and Hydrogen peroxide maintained good activity (inhibition zones of 24 mm, 24 mm, 22 mm, 21 mm). Iodine and Dettol were weak (5 mm, 10 mm). This matches AL-Ratib et al. (2021) for Deco Sept (25 mm), though differs for Cidex OPA and Dettol. Disinfectant efficacy generally increases with contact time, confirming exposure duration as a key factor.

For P. putida, Cidex OPA, Sodium hypochlorite, Hibitane, Deco Sept, and Hydrogen peroxide were effective (28 mm, 25 mm, 22 mm, 21 mm, 21 mm). Dettol and Iodine remained weak (14 mm and 4 mm), suggesting bacterial resistance or insufficient concentrations.

Table 3: Effect of Disinfectants on Pseudomonas Species After 24 Hours

Pseudomonas	Ethanol alcohol	Sodium hypochlorite	Dettol	Deco sept	Cidex OPA	Peroxide hydrogen	Iodine solution	Hibitane
P. aeruginosa	4 mm	22 mm	17 mm	18 mm	25 mm	19 mm	16 mm	19 mm
P. fluorescens	11 mm	24 mm	10 mm	22 mm	24 mm	20 mm	5 mm	21 mm
P. putide	9 mm	25 mm	14 mm	21 mm	28 mm	21 mm	4 mm	22 mm

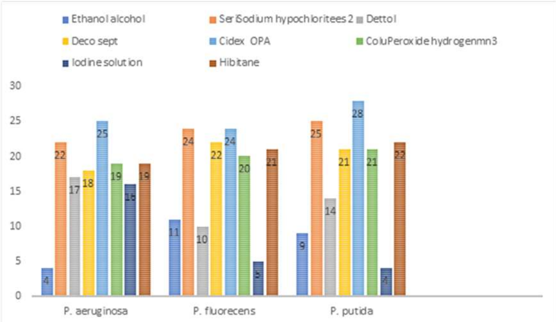


Figure 3: Effect of Disinfectants on Pseudomonas Species After 24 Hours

The highest effects on P. aeruginosa were Cidex OPA (25 mm) and Sodium hypochlorite (22 mm). P. aeruginosa showed high resistance to iodine (12 mm), contrary to AL-Ratib et al. (2021), who reported 18 mm.

P. fluorescens showed highest sensitivity to Cidex OPA and Sodium hypochlorite (24 mm each), but clear resistance to Dettol and Iodine (8 mm, 4 mm).

P. putida showed high sensitivity to the same two disinfectants (28 mm, 25 mm), but resistance to iodine (4 mm).

Ethanol showed weak effects on Pseudomonas species at 24 and 36 hours, attributed to its volatility and incomplete bactericidal effect. Additionally, Pseudomonas can form biofilms—protective layers reducing disinfectant efficacy, including Ethanol. A study showed that 97.5% of P. aeruginosa isolates formed biofilms and exhibited resistance to common disinfectants like Sodium hypochlorite and Ethanol (Mehdi Bakht, et al., 2022).

Table 4: Effect of Disinfectants on Pseudomonas Species After 36 Hours

Pseudomonas	Ethanol alcohol	Sodium hypochlorite	Dettol	Deco sept	Cidex OPA	Peroxide hydrogen	Iodine solution	Hibitane
P. aeruginosa	3 mm	22 mm	15 mm	16 mm	25 mm	19 mm	12 mm	19 mm
P. fluorescens	9 mm	24 mm	8 mm	18 mm	24 mm	20 mm	4 mm	21 mm
P. putide	7 mm	25 mm	13 mm	19 mm	28 mm	21 mm	4 mm	22 mm

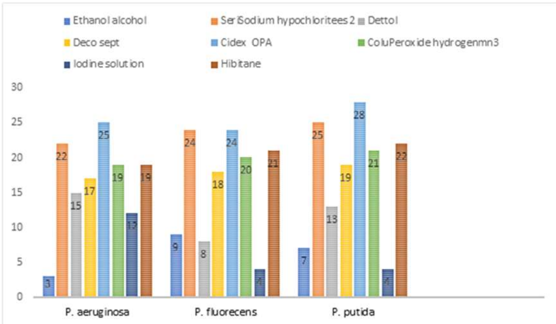
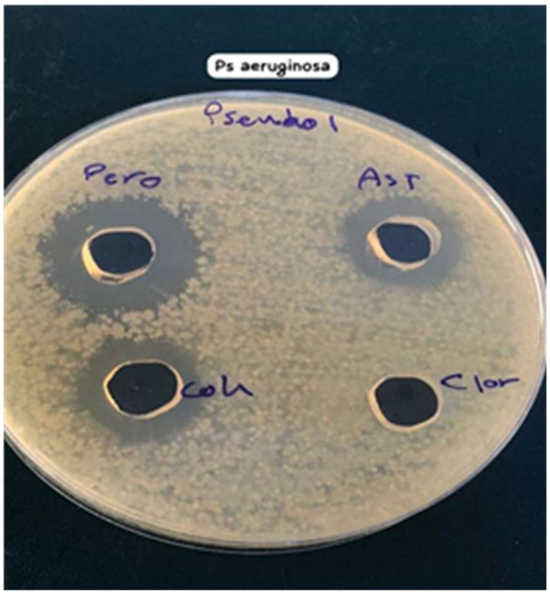
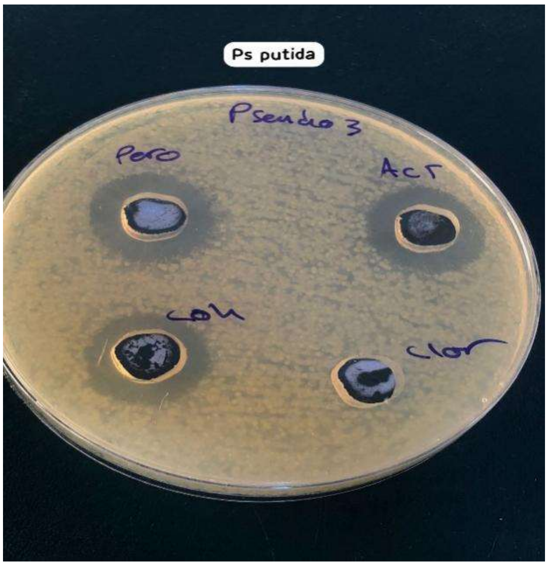
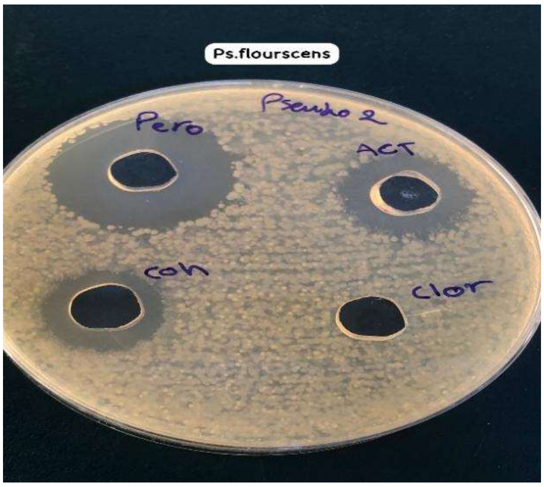
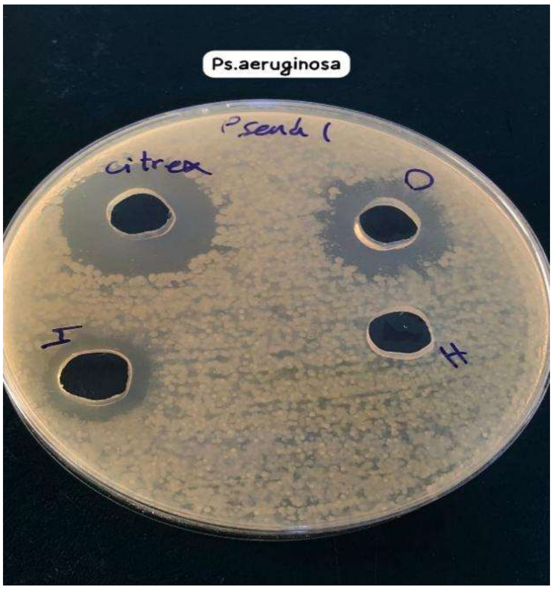


Figure 3: Effect of Disinfectants on Pseudomonas Species after 36 Hours









*** Results and Recommendations**

*** Results**

1- Pseudomonas aeruginosa is the most prevalent bacterium in the environment of Al-Kindi Hospital in Baghdad.

2- The effectiveness of disinfectants against Pseudomonas species varies significantly depending on the type of disinfectant and the contact time between the disinfectant and the pathogenic agents; the longer the contact time, the greater the disinfectant's ability to eliminate the bacteria.

3- The highest efficacy was observed with the disinfectants Cidex OPA and Sodium hypochlorite**,** both showing high effectiveness when used at appropriate concentrations and with sufficient exposure time.

4- The disinfectants Dettol and Iodine showed weak activity against the isolates of Pseudomonas species.

5- Ethanol demonstrated less noticeable activity against Pseudomonas isolates, showing lower efficacy compared to the other disinfectants.

6- Time is a critical factor in the success of the disinfection process.

*** Recommendations**

1- Select disinfectants based on the effectiveness of the active substance against pathogenic microorganisms.

2- Adhere to the specified concentrations and contact times for disinfectants to ensure complete elimination of pathogens.

3- Use chemically different effective agents to prevent the development of acquired microbial resistance.

4- Train staff on proper preparation and dilution techniques for disinfectants to avoid their random or improper use.

5- Conduct periodic evaluations of the effectiveness of the disinfectants in use.

6- The use of disinfectants should comply with proper disinfection protocols in hospitals.

7- Promote research on the resistance mechanisms of Pseudomonas species, focusing on the role of biofilms and efflux systems in reducing disinfectant efficacy.

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