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Evaluation of disinfection methods and effects for handwashing sinks contaminated with *Pseudomonas aeruginosa*

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To address the recurrent contamination of handwashing sinks with *Pseudomonas aeruginosa* (PA) in the Pediatric Intensive Care Unit (PICU), evaluate the PA elimination efficacy of different disinfectants and disinfection methods, and optimize disinfection strategies to prevent PA colonization and transmission, thereby reducing the risk of healthcare-associated infections (HAIs). Nine handwashing sinks in the PICU (August-December 2024) were selected, meeting the criteria of three consecutive positive PA detections, usage frequency ≥ 20 times/day, and no prior special disinfection interventions (excluding those with recent pipe replacement or structural damage). They were divided into three groups (3 sinks per group). Three disinfectants were prepared: 500 mg/L chlorine-containing disinfectant, 0.5% benzalkonium chloride disinfectant, and 75% ethanol stock solution. Three disinfection methods were applied: rinsing method (disinfectant flushing for 1 min twice daily at 5 L/min), soaking method (full coverage soaking with disinfectant for 30 min every morning followed by flushing), and slow-release soaking method (slow injection of disinfectant into the trap for 30-min soaking, 10-min standing, then flushing at 2 L/min). Samples were collected from the overflow outlet, drain, and other sites before disinfection, 1 min after disinfection, and 24 h after disinfection. Colony counting was performed using the pour plate method, PA was identified via the VITEK 2 system and mass spectrometry, and statistical analysis was conducted with SPSS 26.0 ($\alpha = 0.05$). 75% ethanol showed the best immediate effect (colony count < 0.1 CFU/cm^2 and PA detection rate 0% 1 min after disinfection, $P < 0.001$) but PA reoccurred at 24 h. Chlorine-containing disinfectant performed stably (colony count reduced to 0.2 CFU/cm^2, PA detection rate 0%, $P = 0.002$) with the optimal 24-hour bacteriostatic effect. Benzalkonium chloride had weak efficacy (colony count reduced to 5.0 CFU/cm^2, PA detection rate 33.3%). The soaking method and slow-release soaking method were significantly more effective in biofilm removal than the rinsing method (e.g., no PA detected with chlorine-containing disinfectant soaking method and < 10% recurrence rate at 24 h, compared to 66.7% PA positivity rate with the rinsing method at 24 h), and the soaking method was more operable. For PA-contaminated handwashing sinks in the PICU, the chlorine-containing disinfectant soaking method has the best comprehensive effect among the tested methods, combining strong bactericidal power, good long-term bacteriostatic effect, and high operability. It is suitable as a routine disinfection scheme to prevent PA colonization in the ward, providing support for HAI prevention and control. Future research can explore more disinfection methods for ward sinks.

Keywords *Pseudomonas aeruginosa*, Handwashing sink, Healthcare-associated infection, Disinfection

Pseudomonas aeruginosa (PA) is a common clinical Gram-negative bacterium with a single polar flagellum, capable of secreting various virulence factors including exotoxin A, elastase, and phospholipase C[1]. Widely distributed in nature and hospital environments, PA is an opportunistic pathogen with strong environmental adaptability and drug resistance. It can cause HAIs through multiple routes, including direct contact with contaminated surfaces (e.g., sink edges, faucets), inhalation of aerosolized bacteria from splashing water during sink use, indirect transmission via healthcare workers' hands after contact with contaminated sinks, and

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ingestion of water containing PA from compromised plumbing systems, posing a serious threat especially to immunocompromised patients. Globally, the detection rate of carbapenem-resistant *Pseudomonas aeruginosa* (CRPA) has increased from 15% in 2010 to 38% in 2022[2], and infections caused by CRPA have a mortality rate exceeding 20% in the Middle East and American countries[3], PA has ranked among the top five clinically isolated Gram-negative bacteria for ten consecutive years. PA is not only one of the main pathogens of HAIs[4], but also an important pathogen of hospital-acquired lower respiratory tract infections[5]. Its unique metabolic flexibility allows PA to survive in oligotrophic environments, and it can attach to hygiene dead corners such as drains and overflow outlets through biofilm formation, which is difficult to completely remove by routine flushing. For example, biofilms can form in hospital water supply systems, moist surfaces, and gaps in medical equipment[6]. The presence of biofilms not only enhances PA's resistance to disinfectants but also promotes its long-term colonization in hospital environments. In addition, PA's genome has high plasticity, enabling it to quickly acquire drug-resistant genes through horizontal gene transfer, leading to significantly reduced efficacy of broad-spectrum antibiotics such as carbapenems and β-lactams[7]. This evolution of drug resistance not only limits clinical treatment options but also increases the risk of HAI transmission.

In recent years, multiple studies have revealed the key role of handwashing sinks in PA transmission. A French outbreak investigation showed that 75% of PA infection cases in the ICU had consistent genotypes with strains sampled from the environment, and traceability identified the source as biofilm in handwashing sink drains. Similarly, domestic scholars confirmed through whole-genome sequencing that PA clones from an NICU outbreak were highly homologous to strains isolated from handwashing sinks. A multicenter survey of 10 children's hospitals in China found that the PA contamination rate of NICU handwashing sinks was as high as 28.6%, and the homology with strains from infected patients reached 92%[8], indicating the existence of an environment-host transmission chain. These findings highlight the importance of environmental cleaning in infection prevention and control, but optimizing disinfection strategies remains a practical challenge.

A medical institution repeatedly detected PA in handwashing sinks during routine monitoring and formulated a cleaning and disinfection plan using 500 mg/L chlorine-containing disinfectant to flush handwashing sinks twice daily. This plan was implemented for 4 consecutive weeks, but PA was still detected in the water outlets of some handwashing sinks. It was hypothesized that biofilms might exist in the faucet traps, as biofilms can protect bacteria from disinfectant effects—though the institution did not record detailed data on biofilm detection or pipe inspection. Therefore, after reviewing relevant literature, common disinfectants in daily use were utilized to explore handwashing sink disinfection methods, formulate improvement measures, and evaluate the disinfection effect.

Methods

Study objects

Nine handwashing sinks continuously contaminated with PA in the PICU were selected as study objects. Inclusion criteria: ① Positive PA detection in three consecutive samplings; ② Usage frequency ≥ 20 times/day (calculated based on the handwashing frequency of the department); ③ No prior special disinfection interventions. Exclusion criteria: ① Recent pipe system replacement (within 1 week); ② Structural damage or water leakage. The study period was from August to December 2024. Detailed information of the study handwashing sinks is shown in Table S1. All handwashing sinks were distributed in the NICU and general PICU, covering different usage scenarios (e.g., handwashing, instrument cleaning, waste liquid pouring).

Preparation of disinfectants at working concentration

Chlorine-containing disinfectant:1 disinfectant effervescent tablet (containing 250 mg available chlorine) was dissolved in 500 mL distilled water to prepare a working solution with an available chlorine concentration of 500 mg/L, which was prepared fresh before use.

Benzalkonium chloride disinfectant: Benzalkonium chloride stock solution was mixed with distilled water at a ratio of 1:4 according to the instructions to prepare a 0.5% (w/v) disinfectant.

75% ethanol: used directly as stock solution without dilution.

Sampling and detection methods

A 5 cm×5 cm standard sterile template was used. A cotton swab soaked in sterile 0.03 mol/L phosphate buffer or normal saline sampling solution was used to wipe the template area horizontally and vertically 5 times each while rotating the cotton swab. 1–4 template areas were sampled continuously, the part touched by hands was cut off, and the cotton swab was placed into a test tube containing 10 mL sampling solution for inspection.To eliminate the impact of residual disinfectant on sample viability, the sampling swabs were pre-moistened with neutralizing buffer (0.03 mol/L phosphate buffer containing 0.5% sodium thiosulfate for chlorine-containing disinfectants, 0.1% lecithin and 0.5% polysorbate 80 for benzalkonium chloride, and no neutralizer for 75% ethanol).After sampling, the swabs were immersed in 10 mL of the corresponding neutralizing buffer and vortexed for 1 min to ensure complete neutralization of any residual disinfectant. The sampling tube was fully shaken, 1 mL was aspirated with a sterile pipette and inoculated into a petri dish, and 15–20 mL of melted nutrient agar medium (peptone 10 g/L, beef extract 3 g/L, agar 15 g/L, NaCl 5 g/L, pH 7.2–7.4) cooled to 40–45 °C was poured into each dish. Incubation was carried out at 36 °C (± 1 °C) for 48 h, and colony counts were performed.

Sampling time points: before disinfection, 1 min after disinfection, and 24 h after disinfection (to assess residual contamination).

Sampling sites: Focused on the overflow outlet, drain, inner wall of the faucet, and countertop edge (15 cm from the sink edge).

Sampling time	75% Ethanol		0.5% Benzalkonium Chloride		Chlorine-containing disinfectant	
	Colony count (cfu/cm^2)	PA	Colony count(cfu/cm^2)	PA	Colony count(cfu/cm^2)	PA*
Before disinfection	>100	+	>200	+	>100	+
1 min after disinfection	< 0.1	–	0.6	+	0.2	+
24 h after disinfection	60	+	>100	+	1.5	+

Table 1. Rinsing method disinfection effect. *: + Positive for PA, – Negative for PA.

Sampling time	75% Ethanol		0.5% Benzalkonium Chloride		Chlorine-containing disinfectant	
	Colony count(cfu/cm^2)	PA	Colony count(cfu/cm^2)	PA	Colony count(cfu/cm^2)	PA*
Before disinfection	>100	+	>100	+	>100	+
1 min after disinfection	< 0.1	–	5	–	< 0.1	-
24 h after disinfection	10	–	>100	+	5	-

Table 2. Soaking method disinfection effect. *: + Positive for PA, – Negative for PA.

Detection process: Implemented in accordance with the "Hospital Disinfection Sanitary Standard". Colony counting was performed using the pour plate method, and PA identification was verified by both the VITEK 2 system and mass spectrometry.

Statistical analysis: paired t-tests and chi-square tests were performed using SPSS 26.0, with a significance level of $\alpha = 0.05$, and 95% confidence intervals (CI) were calculated.

Disinfection methods

The 9 handwashing sinks were divided into three groups (3 sinks per group), and three disinfection methods were implemented respectively.

The first disinfection method employed was the rinsing method: twice daily (morning and evening), the sink surface and pipes were flushed with the assigned disinfectant for 1 min at a water flow rate of 5 L/min. The second method was the soaking method: every morning, the sink surface and drain were fully covered with the designated disinfectant, soaked for 30 min, then flushed at the same flow rate as the rinsing method. The third method was the slow-release soaking method: disinfectant was slowly injected into the trap through the drain, soaked for 30 min, allowed to stand for 10 min, then the pipes were flushed at a low speed (2 L/min).

Samples of the handwashing sinks were collected after disinfection to evaluate the disinfection effect. During the study period, all sinks underwent standard daily physical cleaning (independent of the experimental disinfection methods) using non-disinfectant neutral detergent (pH 6.5–7.5) followed by rinsing with sterile water. Physical cleaning was performed once daily (07:00–08:00) by ward cleaning staff, focusing on visible debris removal from the sink basin, faucet, and surrounding countertop. No additional physical cleaning or disinfection was conducted outside of this protocol and the experimental interventions.

Results

Among the three disinfection methods, 75% ethanol showed the best immediate disinfection effect within 1 min after disinfection. No PA was detected after disinfection, the colony count decreased from > 100 CFU/cm^2 to < 0.1 CFU/cm^2 within 1 min after disinfection, and the PA detection rate decreased from 100 to 0% ($P < 0.001$); however, the bacteriostatic effect was poor at 24 h after disinfection, and PA reoccurred.

Chlorine-containing disinfectant performed stably under different methods, with a small amount of residue after disinfection but strong bactericidal ability: the colony count decreased from > 100 CFU/cm^2 to 0.2 CFU/cm^2, and the PA detection rate decreased from 100 to 0% ($P = 0.002$), but its 24-hour bacteriostatic effect was the best among the three disinfectants.

Benzalkonium chloride had relatively weak disinfection effect with obvious bacterial residue in some cases, and its performance varied slightly under different methods: the colony count had the smallest decrease (from > 100 CFU/cm^2 to 5.0 CFU/cm^2), and the PA detection rate was still 33.3%.

Details are shown in Tables 1, 2, 3 and 4.

Discussion

The persistent colonization and transmission of *Pseudomonas aeruginosa* (PA) in healthcare settings are attributed to its unique metabolic flexibility and the plasticity of its drug-resistant genes[9]. This bacterium can move via a single polar flagellum to attach to moist surfaces, secreting extracellular polysaccharides (EPS) to form biofilms that significantly enhance its resistance to disinfectants[10]. Biofilms not only provide a physical barrier but also regulate the release of virulence factors (such as elastase and exotoxin A) through quorum sensing, further increasing infection risk. HAIs caused by PA through water-borne facilities such as handwashing sinks have become a global challenge in infection prevention and control. This study systematically evaluated the efficacy

Sampling time	75% Ethanol		0.5% Benzalkonium Chloride		Chlorine-containing disinfectant	
	Colony count(cfu/cm²)	PA	Colony count(cfu/cm²)	PA	Colony count(cfu/cm²)	PA*
Before Disinfection	>100	+	>100	+	>100	+
1 min After Disinfection	< 0.1	–	0.6	–	0.2	–
24 h After Disinfection	< 0.1	–	5	–	5	–

Table 3. Slow-release soaking method disinfection effect. *: + Positive for PA, – Negative for PA.

Disinfectant and disinfection method	Colony count 1 min after disinfection (CFU/cm²)	PA detection*
75% Ethanol		
Rinsing method	<0.1	–
Soaking method	<0.1	–
Slow-release soaking method	<0.1	–
Chlorine-containing disinfectant		
Rinsing method	0.2	+
Soaking method	<0.1	–
Slow-release soaking method	0.2	–
0.5% Benzalkonium Chloride		
Rinsing method	0.6	+
Soaking method	5	–
Slow-release soaking method	0.6	–

Table 4. Comparison of the effects of different disinfectants and methods. *: + Positive for PA, - Negative for PA.

of different disinfectants and disinfection methods in eliminating PA contamination from PICU handwashing sinks, revealing key pathways for optimizing disinfection strategies.

As an increasing number of disinfectant types induce microbial resistance—such as quaternary ammonium salts, biguanides, and chlorhexidine commonly used in healthcare facilities[11], there is an urgent need to identify effective disinfectants and methods to combat pathogenic bacteria in the environment. The mechanisms of bacterial resistance to disinfectants are highly similar to those of antimicrobial resistance, both being related to exposure frequency during use[12–14]. These mechanisms include reduced cell membrane permeability, altered disinfectant targets, plasmid-mediated disinfectant resistance, active efflux pump systems, and the production of specific enzymes to decompose harmful substances[15]. The biofilm matrix of *Pseudomonas aeruginosa* has a dense structure, and EPS in the matrix can affect disinfectant penetration. The main mechanism involves overexpression of the MexAB-OprM drug efflux pump caused by mutations in regulatory genes such as mexR, nalC, or nalD and corresponding amino acids[16], which increases the disinfectant resistance of *Pseudomonas aeruginosa* biofilms[17]. Currently, there are no clear national regulations on the disinfection methods for handwashing sinks. Disinfection methods include chlorine-containing disinfectant wiping and disinfectant wipe wiping, with varying effects[18]. In some outbreak literatures[19], chemical disinfection methods such as 70% ethanol, calcium hypochlorite, or sodium hypochlorite have been reported to achieve good disinfection effects for *Pseudomonas aeruginosa* contamination in water systems, but specific disinfection steps are not described. This study verified the efficacy of three disinfectants and three different disinfection methods in killing PA, providing certain reference value.

75% ethanol exhibited advantages in immediate bactericidal activity in this study. Ethanol exerts its bactericidal effect mainly by disrupting the microbial cell membrane structure[20]. At a concentration of 70–80%, its molecules can quickly penetrate the lipid bilayer of the cell membrane, causing denaturation of membrane proteins, dissolution and rupture of the cell membrane, and leakage of cellular contents such as enzymes and nucleic acids[21]. However, ethanol has poor permeability to biofilms. PA biofilms are encapsulated by a dense matrix composed of EPS, DNA, and proteins, making it difficult for ethanol molecules to penetrate the matrix to reach deep-seated bacteria[22]. In addition, ethanol is highly volatile and cannot provide sustained bacteriostatic effects. Therefore, ethanol is more suitable as an immediate surface disinfectant rather than a long-term biofilm control solution.

Chlorine-containing disinfectants achieve broad-spectrum bactericidal effects through multiple redox reactions[23]. Their active components, hypochlorous acid (HOCl) and hypochlorite (ClO^-), can penetrate the microbial cell wall and act through three pathways: ① Directly oxidize cell membrane lipids and proteins, disrupting membrane permeability; ② Decompose to produce nascent oxygen ([O]), inactivating the enzyme system; ③ Chloride ions alter cellular osmotic pressure leading to lysis[24]. They are classified as high-level disinfectants. Studies have shown that an available chlorine concentration of 500 mg/L can kill common pathogens, but the concentration needs to be increased to 2000–5000 mg/L when facing organic pollution or Clostridioides difficile spores[25]. Novel complex chlorine technology can slowly release hypochlorous acid,

significantly reducing corrosion to metal instruments while maintaining bactericidal power[26]. In this study, the bactericidal effect of the chlorine-containing disinfectant soaking method (30 min) was significantly superior to the rinsing method (1 min), confirming its time-dependent characteristics. The oxidative effect of hypochlorous acid on EPS can gradually break down the biofilm structure, but sufficient contact time (> 15 min) is required to achieve deep penetration. It is worth noting that high-concentration chlorine-containing disinfectants (> 1000 mg/L) may corrode metal pipes[27], requiring a balance between bactericidal effect and equipment safety. In addition, long-term use of chlorine-containing disinfectants may induce the expression of disinfectant resistance genes (such as qacEΔ1) in PA, increasing the risk of disinfection failure[28].

Benzalkonium chloride, as a cationic surfactant, kills bacteria by disrupting cell membrane potential and osmotic balance. However, its effect on PA biofilms is limited due to: ① EPS carries a negative charge, which can adsorb quaternary ammonium salt cations, reducing the effective bactericidal concentration; ② PA's efflux pump system (such as MexAB-OprM) can actively pump out quaternary ammonium salt molecules, reducing intracellular drug accumulation[29]. In this study, the PA detection rate in the benzalkonium chloride group was still 33.3% after disinfection, consistent with the reported trend of quaternary ammonium salt resistance[30]. In recent years, the detection rate of plasmid-mediated quaternary ammonium salt resistance genes (such as qacA/B) in PA has increased[31], indicating that caution should be exercised when selecting such disinfectants.

Sterilization rate is positively correlated with sterilization time and dosage. Insufficient disinfectant dosage can lead to incomplete disinfection or even disinfection failure, inducing bacterial resistance to disinfectants and increasing the risk of HAIs[32]. The traditional rinsing method relies on mechanical flushing with water flow, which can remove surface planktonic bacteria but is ineffective against hidden areas (such as overflow outlets and traps) and biofilms. This study showed that the PA positivity rate rebounded to 66.7% 24 h after disinfection with the rinsing method. Improvement directions include: ① Increasing the rinsing frequency to 4 times daily; ② Using pulsed high-pressure water flow (> 50 psi) to disrupt biofilm adhesion; ③ Combining with pipeline endoscopy for physical debridement. However, high-frequency rinsing may accelerate pipeline aging, requiring cost-benefit analysis. The soaking method significantly improves biofilm removal efficiency by extending disinfectant contact time (30 min). In this study, no PA was detected after disinfection in the chlorine-containing disinfectant soaking group, and the 24-hour recurrence rate was less than 10%. In practical operation, an automated perfusion system can be used to inject disinfectant into the handwashing sink and close the drain to ensure full coverage of complex structures. In addition, after soaking, flushing with low-speed water flow (2 L/min) is required to avoid reducing the effect by dispersing the disinfectant with high-speed water flow.

From a practical perspective, the chlorine-containing disinfectant soaking method (30 min daily) has minimal disruption to workflow. Sinks were taken out of operation during the early morning (06:00–06:30), a period of low usage in the PICU/NICU. The 500 mg/L chlorine-containing disinfectant produces negligible fumes and odor when used as directed, and no complaints from staff or adverse effects on patients were reported during the study. The rinsing method (1 min twice daily) has no workflow disruption but is less effective, while the slow-release soaking method requires specialized equipment for slow injection, increasing operational complexity.

The trap is the core colonization area for PA biofilms. The slow-release soaking method designed in this study maintains a disinfectant concentration of ≥ 500 mg/L in the trap for 30 min by slowly injecting disinfectant (10 min), effectively removing deep biofilms. This method is complementary to the principle of the electronic oscillation disinfector invented by de Jonge et al.[33] which disrupts the biofilm structure through high-frequency sound waves but has a high cost. The slow-release soaking method achieves similar effects at low cost, especially suitable for hospitals in low- and middle-income countries with limited resources.

Structural defects of handwashing sinks are one of the root causes of PA contamination. Current handwashing sinks mostly adopt fixed drain pipes and shallow basin designs, which are prone to water splashing (splash radius up to 1.5 m) and biofilm accumulation. A multicenter study in the Netherlands showed that improved handwashing sink designs can reduce the PA contamination rate by 76%. Su Jing et al.[34] found that continuous soaking with 1 ~ 2 mg/L polyiodide resin-filtered water or brominated polystyrene hydantoin resin filtered water is more effective in removing viable and dead bacteria in biofilms and biofilm matrices, and can be used in the field of biofilm control in water systems. de Jonge et al.[33]invented an electronic oscillation disinfector for trap disinfection. Installed at the trap of ICU sinks during non-outbreak periods, it can effectively reduce the colonization rate of *Pseudomonas aeruginosa* in sinks from 51 to 46%. Medical institutions can take the following measures to improve handwashing sinks according to their own conditions: adopting detachable traps for monthly cleaning and disinfection; installing splash guards or deep basin designs (depth > 20 cm); using antibacterial coating materials (such as silver ion-containing ceramics) to inhibit biofilm formation.

Alternative strategies to reduce sink-related PA transmission include reducing the number of non-essential sinks in healthcare settings and promoting alcohol-based hand rub (ABHR) as a primary hand hygiene method, as ABHR effectively kills PA and reduces reliance on sink use. However, sinks remain essential for tasks such as instrument cleaning and waste liquid disposal, so optimizing sink disinfection (as demonstrated in this study) remains a critical complement to these strategies. Future research could explore the combined effect of sink reduction and targeted disinfection on HAIs.

This study has several limitations. First, the sample size of nine sinks is relatively small, which may limit the generalizability of the results. Future studies with larger sample sizes across multiple healthcare facilities are needed to validate the findings. Second, the study was conducted in a single institution's PICU and NICU, so results may not be directly applicable to other wards (e.g., general wards, surgical ICUs) with different usage patterns or environmental conditions. Third, we did not evaluate the long-term corrosion effect of chlorine-containing disinfectants on pipe materials, which warrants further investigation.

In summary, this study evaluated the disinfection efficacy of three disinfectants and three disinfection methods for handwashing sinks. 75% ethanol had the best immediate disinfection effect, chlorine-containing disinfectant performed stably, while benzalkonium chloride had relatively weak disinfection effect. The

soaking method was more effective in removing residual *Pseudomonas aeruginosa* than the rinsing method. The soaking method is more operable than the slow-release soaking method. It is recommended to use the chlorine-containing disinfectant soaking method for routine disinfection to prevent *Pseudomonas aeruginosa* colonization in the ward. Future research can explore more cleaning and disinfection methods for ward sinks.

Data availability

The data that support the findings of this study are derived from the routine hospital infection monitoring records and experimental data of the Maternal and Child Health Hospital of Guangxi Zhuang Autonomous Region. Due to the sensitivity of these data (which involve hospital-specific environmental monitoring information and clinical site-related records), restrictions apply to the availability of the raw data, and they are not publicly accessible.However, de-identified data necessary to replicate the study's key findings are available from the corresponding author (Li Yan, E-mails: 260291278@qq.com; angelliyan2008@yeah.net) upon reasonable request, provided that such requests are accompanied by a formal application and with the approval of the Medical Research Ethics Committee and the Department of Hospital Infection Control of the Maternal and Child Health Hospital of Guangxi Zhuang Autonomous Region. All materials (including disinfectant specifications, sampling templates, and detection kits) used in this study are commercially available and do not involve proprietary or restricted materials.

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References

1. Zhang, X. et al. A comprehensive review of the pathogenic mechanisms of *Pseudomonas aeruginosa*: Synergistic effects of virulence factors, quorum sensing, and biofilm formation. *Front. Microbiol.***16**, 1619626 (2025).
2. Zhu, Y. et al. The epidemiology and virulence of carbapenem-resistant *Pseudomonas aeruginosa* in China. *Lancet Microbe.* **4** (9), e665 (2023).
3. Reyes, J. et al. Global epidemiology and clinical outcomes of carbapenem-resistant *Pseudomonas aeruginosa* and associated carbapenemases (POP): A prospective cohort study. *Lancet Microbe*. **4** (3), e159–e170 (2023).
4. Reynolds, D. & Kollef, M. The epidemiology and pathogenesis and treatment of *Pseudomonas aeruginosa* infections: An update. *Drugs* **81** (18), 2117–2131 (2021).
5. Li, Y. et al. The global epidemiology of ventilator-associated pneumonia caused by multi-drug resistant *Pseudomonas aeruginosa*: A systematic review and meta-analysis. *Int. J. Infect. Dis.* **139**, 78–85 (2024). 2024-02-01.
6. Letizia, M., Diggle, S. P. & Whiteley, M. *Pseudomonas aeruginosa*: Ecology, evolution, pathogenesis and antimicrobial susceptibility. *Nat. Rev. Microbiol.* **23** (11), 701–717 (2025).
7. Gholipour, S., Nikaeen, M., Mehdipour, M., Mohammadi, F. & Rabbani, D. Occurrence of chlorine-resistant *Pseudomonas aeruginosa* in hospital water systems: Threat of waterborne infections for patients. *Antimicrob. Resist. Infect. Control*. **13** (1), 111 (2024).
8. Tomczyk, S. et al. Control of carbapenem-resistant enterobacteriaceae, acinetobacter baumannii, and *Pseudomonas aeruginosa* in healthcare facilities: A systematic review and reanalysis of quasi-experimental studies. *Clin. Infect. Dis.* **68** (5), 873–884 (2019).
9. Cortes-Lara, S., Barrio-Tofino, E. D., Lopez-Causape, C. & Oliver, A. Predicting *Pseudomonas aeruginosa* susceptibility phenotypes from whole genome sequence resistome analysis. *Clin. Microbiol. Infec.* **27** (11), 1631–1637 (2021).
10. Jaillard, M. et al. Correlation between phenotypic antibiotic susceptibility and the resistome in *Pseudomonas aeruginosa*. *Int. J. Antimicrob. AG.* **50** (2), 210–218 (2017).
11. Maillard, J. Y. & Pascoe, M. Disinfectants and antiseptics: Mechanisms of action and resistance. *Nat. Rev. Microbiol.* **22** (1), 4–17 (2024).
12. Fernandes, A. R., Rodrigues, A. G. & Cobrado, L. Effect of prolonged exposure to disinfectants in the antimicrobial resistance profile of relevant micro-organisms: A systematic review. *J. Hosp. Infect.* **151**, 45–59 (2024).
13. Rozman, U., Pusnik, M., Kmetec, S., Duh, D. & Sostar, T. S. Reduced susceptibility and increased resistance of bacteria against disinfectants: A systematic review. *Microorganisms* **9** (12), 2550 (2021).
14. Schrader, S. M., Vaubourgeix, J. & Nathan, C. Biology of antimicrobial resistance and approaches to combat it. *Sci. Transl. Med.* **12** (549), eaaz6992. (2020).
15. Liu, Y. et al. Regulatory mechanisms of quorum sensing system of bacteria in response to chlorine and ozone disinfection. *Environ. Sci. Technol.* **58** (30), 13337–13347 (2024) .
16. Amsalu, A. et al. Efflux pump-driven antibiotic and biocide cross-resistance in *Pseudomonas aeruginosa* isolated from different ecological niches: A case study in the development of multidrug resistance in environmental hotspots. *Microorganisms* **8** (11), 1647 (2020).
17. Sigler Zekanović, M., Begić, G., Medić, A., Gobin, I. & Tomić Linšak, D. Effects of a combined disinfection method on *Pseudomonas aeruginosa* biofilm in freshwater swimming pool. *Environments* **8** (9), 103 (2022).
18. Zhao, Y. et al. Efficacy of 75% alcohol in pretreatment of the Andersen sampler in trapping maximum airborne microbes. *Aerobiologia* **37**(1), 171–178 (2021).
19. Decraene, V. et al. An outbreak of multidrug-resistant *Pseudomonas aeruginosa* in a burns service in the North of England: Challenges of infection prevention and control in a complex setting. *J. Hosp. Infect.* **100** (4), e239–e245 (2018).
20. Fu, L. et al. Different efficacies of common disinfection methods against Candida auris and other Candida species. *J. Infect. Public. Health* **13** (5), 730–736 (2020).
21. Ramirez, A. et al. Surface contamination in the operating room: Use of adenosine triphosphate monitoring. *J. Anesth.* **33** (1), 85–89 (2019).
22. Rostamani, M. et al. Phenotypic and genotypic determination of resistance to common disinfectants among strains of acinetobacter baumannii producing and non-producing biofilm isolated from Iran. *BMC Microbiol.* **24** (1), 323 (2024).
23. Ahmed, R. & Ahmed, S. Exploring the efficacy of hypochlorous acid as a cost effective environmental decontaminant in dentistry: A scoping review. *Niger J. Clin. Pract.* **28** (4), 425–445 (2025).
24. Li, J. et al. The effect of different concentrations of chlorine-containing disinfectants on high-frequency contact table in intensive care units: A quasi-experimental study. *PLOS ONE*. **18** (2), e281802 (2023).
25. Alkhodary, M. A. Effect of controlled surface roughness and biomimetic coating on titanium implants adhesion to the bone: An experiment animal study. *Saudi Dent. J.* **35** (7), 819–826 (2023).
26. Cao, K. F. et al. Modeling and optimization of synergistic ozone-ultraviolet-chlorine process for reclaimed water disinfection: From laboratory tests to software simulation. *Water Res.* **243**, 120373 (2023).

27. Li, Z. et al. Management and microbial monitoring of final rinse water for flexible endoscopes in 290 hospitals in Jiangsu Province, China: A multicenter cross-sectional study. *Antimicrob. Resist. Infect. Control.* **14** (1), 40 (2025).
28. Wang, H. M. et al. Detection and analysis of drug and disinfectant resistance genes in the sewage of a center for disease control and prevention. *Infect. Drug Resist.* **18**, 2079–2086 (2025).
29. Liu, Z. et al. Surface-mediated fluorescent sensor array for identification of gut microbiota and monitoring of colorectal cancer. *Talanta* **274**, 126081 (2024).
30. Soltani, M., Ravine, T. J. & Davis, J. J. Novel boronium salt exhibits substantial antibacterial activity when compared to a commercial quaternary ammonium disinfectant. *Bioorg. Med. Chem. Lett.* **36**, 127808 (2021).
31. Shao, D. et al. Quaternary ammonium compounds: A new driver and hidden threat for mcr-1 prevalence in hospital wastewater and human feces. *Environ. Sci. Technol.* **59** (3), 1565–1576 (2025).
32. LaBreck, P. T. et al. Systematic analysis of efflux pump-mediated antiseptic resistance in *Staphylococcus aureus* suggests a need for greater antiseptic stewardship. *Msphere* **5** (1), 10 (2020).
33. de Jonge, E., de Boer, M., van Essen, E., Dogterom-Ballering, H. & Veldkamp, K. E. Effects of a disinfection device on colonization of sink drains and patients during a prolonged outbreak of multidrug-resistant *Pseudomonas aeruginosa* in an intensive care unit. *J. Hosp. Infect.* **102** (1), 70–74 (2019).
34. Ruixue, H. & Jing, S. Removal effect of disinfection factors in low concentration on *Pseudomonas aeruginosa* biofilm. *Chin. J. Prev. Med.* **10** (57), 1620–1624 (2023).

Author contributions

Authors' ContributionsLD (Liao Dan) put forward the core research design and analysis ideas, and was the main contributor to drafting the manuscript. WYN (Wei Yanni), LGY (Liao Guiyi), LWT (Liang Wenting), and MZJ (Ma Zhangjie) participated in sample collection, data collation, and disinfection operation implementation, providing key experimental data for the study. QYM (Qiu Yanmei) assisted in microbial detection and colony counting, ensuring the accuracy of experimental results.LY (Li Yan, corresponding author) guided the overall research process, participated in data analysis and interpretation, and made important contributions to manuscript revision and finalization. LD and LY jointly completed the statistical analysis of data using SPSS 26.0. All authors read, reviewed, and approved the final version of the manuscript.

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Declarations

Competing interests

The authors declare no competing interests.

Ethical approval

Not applicable. This study only involves environmental sampling of handwashing sinks in the Pediatric Intensive Care Unit (PICU) of the Maternal and Child Health Hospital of Guangxi Zhuang Autonomous Region, without involving human subjects, collection of personal health information, or intervention on patients. The sampling process strictly adheres to the hospital's routine infection control monitoring protocols and relevant operational standards, and does not require ethical approval.

Additional information

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