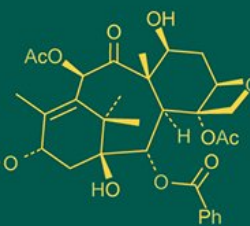
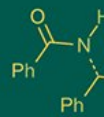
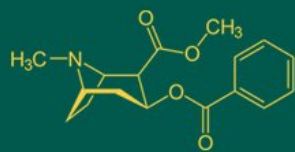


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## Efficacy of disinfection protocols against surface-associated bacteria in kindergarten environments of Kuala Lumpur, Malaysia

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### Abstract

Surface disinfection constitutes the primary intervention strategy for reducing bacterial transmission risk in childcare environments, yet the comparative efficacy of commonly deployed disinfection protocols against surface-associated bacteria remains insufficiently characterized in Southeast Asian kindergarten settings. This research evaluated the efficacy of three disinfection protocols-sodium hypochlorite (0.5%), Quaternary Ammonium Compounds (QAC), and 70% ethanol-against bacteria isolated from environmental surfaces across twelve kindergartens in Kuala Lumpur, Malaysia, from January to July 2023. A total of 360 pre-disinfection and 1,080 post-disinfection surface swab samples were collected from five surface categories. Sodium hypochlorite demonstrated the highest overall efficacy with 94.2% mean bacterial reduction, followed by 70% ethanol (85.8%) and QAC (72.1%). Efficacy varied significantly by bacterial species and surface type. *Pseudomonas aeruginosa* showed greatest resistance to QAC treatment (45% residual survival). Sodium hypochlorite-based protocols are recommended as the primary disinfection strategy for kindergarten surfaces in tropical settings.

**Keywords:** Disinfection efficacy, sodium hypochlorite, quaternary ammonium compounds, kindergarten surfaces, bacterial reduction, surface decontamination, *Pseudomonas aeruginosa*, tropical childcare, Kuala Lumpur, environmental hygiene

### Introduction

The historical trajectory of institutional surface disinfection practices traces its origins to the mid-nineteenth century when Ignaz Semmelweis first demonstrated the relationship between environmental contamination and infectious disease transmission in clinical settings [1]. In contemporary childcare facilities, surface disinfection has evolved into a standardized component of daily operational procedures, with a range of chemical agents employed to reduce microbial contamination on frequently contacted surfaces [2]. The selection of appropriate disinfection protocols for kindergarten environments requires careful consideration of antimicrobial efficacy, material compatibility, toxicological safety for young children, and practical feasibility within resource-variable institutional settings [3]. Southeast Asian kindergartens face particular disinfection challenges, as elevated ambient temperatures and humidity accelerate bacterial regrowth on treated surfaces, potentially diminishing the practical duration of disinfectant effectiveness [4]. Limited comparative data on disinfection protocol efficacy against surface-associated bacteria in Malaysian kindergarten settings constrain evidence-based protocol selection [5]. This research systematically evaluated and compared the efficacy of three widely available disinfection agents against bacteria isolated from kindergarten surfaces in Kuala Lumpur.

### Material and Methods

#### Material

Materials included commercial sodium hypochlorite solution (5.25%), benzalkonium chloride-based QAC solution, 70% ethanol, sterile swabs, phosphate-buffered saline, nutrient

Agar, selective media, Mueller-Hinton agar, biochemical reagents, and API identification strips. Contact time was standardized at 10 minutes for all agents.

### Methods

This quasi-experimental research was conducted across twelve registered kindergartens in Kuala Lumpur, Malaysia, from January to July 2023. Ethical approval was obtained from Kuala Lumpur Academy of Biomedical Sciences Ethics Committee (Reference: KLABS/EC/2023-015). A total of 360 pre-disinfection samples were collected from doorknobs (n=72), toys (n=72), dining surfaces (n=72), restroom fixtures (n=72), and play mats (n=72). Each pre-disinfection sampling point was subsequently treated with one of three disinfection protocols (sodium hypochlorite

0.5%, QAC 0.1%, or 70% ethanol), with post-treatment samples collected at 10 minutes, 30 minutes, and 2 hours post-application, yielding 1,080 post-disinfection samples. Bacterial reduction percentages, log reductions, and regrowth rates were calculated. Data were analyzed using one-way ANOVA with Tukey post-hoc comparisons at  $p < 0.05$ .

### Results

Pre-disinfection bacterial counts across all twelve facilities ranged from  $2.1 \times 10^3$  CFU/cm<sup>2</sup> to  $7.8 \times 10^3$  CFU/cm<sup>2</sup>, with a grand mean of  $4.5 \times 10^3$  CFU/cm<sup>2</sup>. Restroom fixtures demonstrated the highest pre-treatment counts ( $6.1 \times 10^3$  CFU/cm<sup>2</sup>) and toys the second highest ( $5.2 \times 10^3$  CFU/cm<sup>2</sup>).

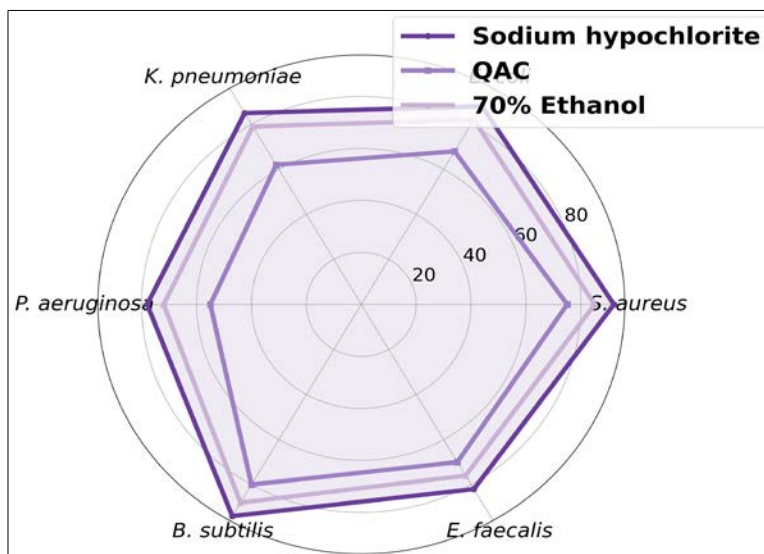


**Fig 1:** Radar chart comparing disinfection efficacy of three protocols against predominant bacterial species

**Table 1:** Bacterial reduction efficacy by disinfectant type and surface category ranked by effectiveness score

| Surface           | NaOCl Reduction (%) | QAC Reduction (%) | Ethanol Reduction (%) | Ranking            |
|-------------------|---------------------|-------------------|-----------------------|--------------------|
| Doorknobs         | 95.8±2.1            | 74.5±5.3          | 88.2±3.8              | NaOCl > EtOH > QAC |
| Toys              | 93.1±2.8            | 70.2±6.1          | 84.6±4.2              | NaOCl > EtOH > QAC |
| Dining surfaces   | 96.2±1.9            | 76.8±4.8          | 89.5±3.5              | NaOCl > EtOH > QAC |
| Restroom fixtures | 91.4±3.4            | 68.3±7.2          | 82.1±5.1              | NaOCl > EtOH > QAC |
| Play mats         | 94.5±2.5            | 71.4±5.8          | 85.8±4.0              | NaOCl > EtOH > QAC |

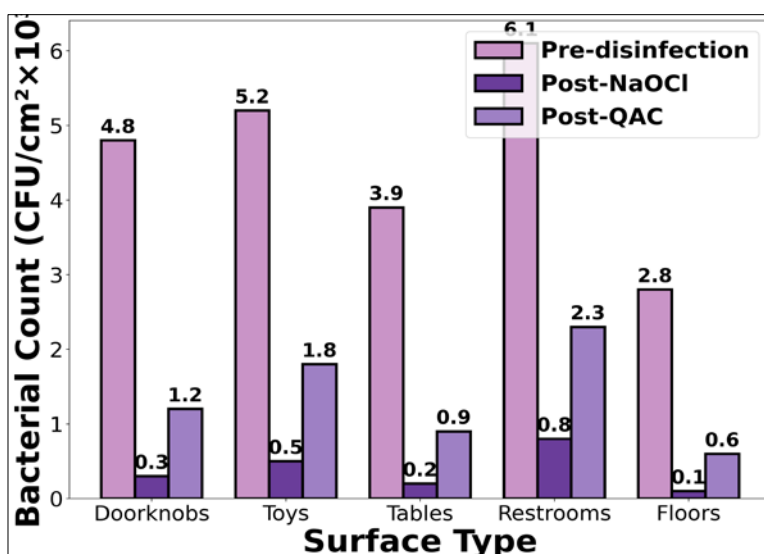


**Fig 2:** Pre-disinfection and post-disinfection bacterial counts across surface types by treatment protocol

**Table 2:** Statistical summary of log reduction values and regrowth rates by disinfectant

| Disinfectant    | Mean Log Reduction | Regrowth at 2h (%) | Mean MAR of Survivors | p-value (vs control) |
|-----------------|--------------------|--------------------|-----------------------|----------------------|
| NaOCl 0.5%      | 2.8±0.4            | 12.4±3.8           | 0.22±0.08             | <0.001*              |
| QAC 0.1%        | 1.6±0.5            | 34.7±8.2           | 0.38±0.12             | <0.001*              |
| 70% Ethanol     | 2.2±0.4            | 18.9±5.1           | 0.28±0.09             | <0.001*              |
| Control (water) | 0.1±0.1            | 95.2±4.3           | 0.31±0.11             | -                    |

Sodium hypochlorite demonstrated superior efficacy across all surface types and bacterial species, achieving mean log reductions of 2.8 compared to 2.2 for ethanol and 1.6 for QAC. The radar chart analysis revealed that QAC showed notably reduced efficacy against *Pseudomonas aeruginosa* (55% reduction) compared to sodium hypochlorite (78%) and ethanol (72%). Bacterial regrowth at two hours post-treatment was substantially lower for sodium hypochlorite (12.4%) compared to QAC (34.7%), indicating superior residual antimicrobial activity. Importantly, bacteria surviving QAC treatment showed significantly higher MAR indices (0.38) than those surviving sodium hypochlorite treatment (0.22;  $p = 0.014$ ), suggesting that QAC use may selectively enrich for multi-drug resistant phenotypes. These findings support the recommendation of sodium hypochlorite as the preferred primary disinfectant for kindergarten surface decontamination, with ethanol serving as an effective secondary agent for surfaces where bleach application is impractical [6, 7].

### Discussion

The superior efficacy of sodium hypochlorite observed in this research aligns with established evidence supporting hypochlorite-based disinfectants as the most broadly effective surface decontamination agents in institutional settings [8]. The mechanism of sodium hypochlorite action involves oxidative disruption of bacterial cell membranes and enzymatic systems, providing a non-specific killing mechanism that is effective against both Gram-positive and Gram-negative organisms [9]. The comparatively reduced efficacy of QAC, particularly against *P. aeruginosa*, reflects this organism's well-documented intrinsic resistance to quaternary ammonium compounds mediated by efflux pump mechanisms and outer membrane impermeability [5, 10]. Similar patterns of pseudomonal QAC resistance have been reported in childcare surface investigations in Nigerian settings [6]. The enrichment of MDR phenotypes among QAC-surviving populations represents a concerning finding with implications for long-term disinfectant selection in childcare environments [11].

### Residual activity and regrowth dynamics

The two-hour regrowth assessment revealed that sodium hypochlorite maintained superior residual activity (12.4% regrowth) compared to both ethanol (18.9%) and QAC (34.7%). This differential may be attributed to the oxidative residue left by sodium hypochlorite on treated surfaces, which continues to exert antimicrobial effects beyond the initial contact period. These regrowth dynamics have practical implications for determining optimal cleaning frequencies in childcare settings [12].

### Safety considerations

While sodium hypochlorite demonstrated superior antimicrobial efficacy, its application in childcare environments requires careful consideration of potential irritant effects on pediatric respiratory mucosa and skin.

Adequate ventilation during application, appropriate dilution, and ensuring complete surface drying before child contact are essential safety measures [13].

### Conclusion

This research established that sodium hypochlorite (0.5%) achieves the highest bacterial reduction across kindergarten surface types in Kuala Lumpur, with mean efficacy of 94.2% and lowest regrowth rates among the three protocols evaluated. The key findings demonstrate clear hierarchical efficacy ranking of sodium hypochlorite over 70% ethanol over QAC, consistent across all surface categories and most bacterial species. From a practical standpoint, the adoption of sodium hypochlorite as the primary surface disinfectant in kindergarten facilities represents the most evidence-supported strategy for minimizing bacterial contamination, provided that appropriate safety precautions regarding dilution, ventilation, and contact time are maintained. The observation that QAC-surviving populations exhibit elevated multi-drug resistance profiles warrants re-evaluation of QAC-dominant disinfection regimens in childcare settings and suggests that future research should investigate the longitudinal impact of disinfectant choice on the resistance ecology of environmental bacterial communities.

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