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Research Article

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New commercial wipes inhibit the dispersion and adhesion of *Staphylococcus aureus* and *Pseudomonas aeruginosa* biofilms

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Abstract

Aim: Bacterial biofilms can form on surfaces in hospitals, clinics, farms, and food processing plants, representing a possible source of infections and cross-contamination. This study investigates the effectiveness of new commercial wipes against *Staphylococcus aureus* and *Pseudomonas aeruginosa* biofilms (early attachment and formed biofilms), assessing LH SALVIETTE wipes (Lombarda H S.r.l.) potential for controlling biofilm formation.

Methods and results: The wipes efficacy was studied against the early attachment phase and formed biofilm of *S. aureus* ATCC 6538 and *P. aeruginosa* ATCC 15442 on a polyvinyl chloride (PVC) surface, following a modified standard test EN 16615:2015, measuring Log_{10} reduction and cell viability using live/dead staining. It was also evaluated the wipes anti-adhesive activity over time (3 h, 2 4h), calculating $CFU.mL^{-1}$ reduction. Data were analyzed using *t*-student test. The wipes significantly reduced both early phase and formed *S. aureus* biofilm, preventing dispersion on PVC surfaces. Live/dead imaging showed bacterial cluster disaggregation and killing action. The bacterial adhesive capability decreased after short-time treatment (3 h) with the wipes compared to 24 h.

Conclusions: Results demonstrated decreased bacterial count on PVC surface both for early attachment phase and formed biofilms, also preventing the bacterial biofilm dispersion.

Impact Statement

This data emphasizes the effectiveness of disinfection wipes repeated application on surfaces in healthcare facilities, farms, and food industry to prevent biofilm formation, colonization, and cross-contamination. Considering the fundamental role of prevention in the One Health approach, the suggestion to use disinfecting wipes could help to control sessile microbial proliferation, avoiding potential contamination.

Keywords: *Staphylococcus aureus*; *Pseudomonas aeruginosa*; antimicrobial wipes; hospital acquired infections; surface disinfection; antibiofilm activity

Introduction

Staphylococcus aureus (*S. aureus*) and *Pseudomonas aeruginosa* (*P. aeruginosa*) are common and important bacterial pathogens, and are isolated from several infection sites of both humans and animals (Heaton et al. 2020, Abd El-Ghany 2021). *Staphylococcus aureus* colonizes the skin, the nose, and the mucosal surfaces, and the carriage of *S. aureus* is considered as risk factor for subsequent infections, especially in surgical patients, patients on hemodialysis, and patients in intensive care units (Howden et al. 2023). In addition, *S. aureus* affects human and animal hosts and can cause multiple diseases. In humans, *S. aureus* is responsible for several infections such as skin infections, bacteremia, staphylococcal foodborne poisoning (SFP), and osteomyelitis. In animals, *S. aureus* is a prevalent pathogen that infects cattle and causes livestock production-related diseases with different severity. For instance, *S. aureus* is accountable for bovine mastitis in dairy cattle, exudative dermatitis in pigs, and wound infections (Pal et al. 2023). SFP is documented with *S. aureus*-contaminated food, e.g. meat, egg, and unpasteurized milk products (Bencardino et al. 2021). Interestingly, *S. aureus* showed resistance against various antibiotics and the *S. aureus* persister cells can withstand the stress entering into a state of dormancy that provides tolerance against high concentrations of antibiotics (Cheung et al. 2021, Pan et al. 2023).

The opportunistic pathogen, *P. aeruginosa* can cause hospital-acquired infections (HAIs), especially in the burn wounds, immunocompromised patients, and cystic fibrosis patients, and it has the ability to infect animals causing otitis, corneal ulcers, urinary tract, and soft tissue infections with the possibility to transfer to humans (Santaniello et al. 2020, Abd El-Ghany et al. 2021). Moreover, *P. aeruginosa* was isolated from different food products such as milk, meat, and several types of fruits and vegetables (Li et al. 2023). *Pseudomonas aeruginosa* is well known for its antimicrobial resistance, the ability to form biofilms, and persistence in the

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Table 1. Formulations of RTU "LH SALVIETTE®" wipes according to the manufacturers.

Formulation in 100 g solution	Organic load	Contact time
4.00 g Isopropanol 0.50 g didecyl-dimethyl ammonium chloride 0.25 g o-phenyl phenol 0.025 g fatty alcohol ethoxylates 0.01 g Monoethanolamine	3 g.L^{-1} bovine albumin solution + 3 mL.L^{-1} sheep erythrocytes (simulates high organic burden)	60 s

hospital environments. This bacterium showed significant involvement in the device-associated nosocomial infections, including ventilator-associated pneumonia and catheter-related bloodstream infections (Labovská, S. 2021).

Both bacteria can withstand the stressful environment by forming mono/polymicrobial biofilms. This step takes place by the bacterial phenotypical change from the free-living planktonic state to the sessile state, by the attachment to biotic/abiotic surfaces, and building complex and interactive community embedded in extracellular polymeric substance (EPS). EPS is responsible for preserving the biofilm integrity and promoting the antibacterial tolerance to antibiotics and biocides (Yung et al. 2021). Bacterial biofilm plays an important role in the survival of the pathogens leading to extended colonization, which increases the risk to the patients and healthcare workers. The most common bacterial strains responsible for the HAIs are *Streptococcus* spp., *Acinetobacter* spp., enterococci, *P. aeruginosa*, coagulase-negative staphylococci, *S. aureus*, and *Escherichia coli* (Isigi et al. 2023).

In addition to the pathogens, the medical devices are considered important in the spread of nosocomial infections. They are divided into critical, semi-critical and non-critical items. Each type needs to be in a certain level of disinfection or sterility depending on its contact with the sterile tissue of the human body, mucous membranes, non-intact skin, and intact skin. While critical devices require sterility, other devices may need high or low level of disinfection (Rutala and Weber 2021).

In order to achieve the required level of disinfection, several biocides are used in this process, including alcohols, phenolic compounds, hypochlorite, quaternary ammonium compounds (QAC), chlorhexidine, proxygenes, and aldehydes. They act in different mechanisms of action to affect the microbial cell (Bharti et al. 2022).

Nonetheless, some bacterial strains can resist the disinfectants activity, creating a serious issue in the health care facilities (Rozman et al. 2021). Based on the importance of maintaining specific disinfection criteria, the European Standard tests provide a several tests to evaluate the efficacy of different disinfection products applied on instruments/surfaces, taking into consideration the application requirements regarding the medium organic matter contamination simulation and the application time. The European standard EN 16615:2015 test (4-field test method) aimed to evaluate the bactericidal and yeasticidal activity of disinfectant wipes on non-porous surfaces with mechanical force application. The 4-field test method was evaluated for its activity, reproducibility, and repeatability, and it showed a reliable results in comparison to an automated method, making it a recommended to test wipes efficacy (Jacobshagen et al. 2020). This European standard EN 16615:2015 was applied in several studies to study the activity against planktonic microbial cells (Tarka et al. 2019, Jacobshagen et al. 2020, Müller et al. 2020, Tyski et al. 2021), and it was modified to conform to spores (Gemein et al. 2019, Gemein et al. 2022, Verguet et al. 2023) and viruses (Jahromi et al. 2020).

This study aimed to utilize the previously mentioned standard test to evaluate the anti-dispersion and anti-adhesive action of commercial disinfectant wipes against the early phase attachment of bacteria and formed biofilms of *S. aureus* and *P. aeruginosa* on PVC surface. The importance of this study is related to the frequent isolation of these two bacteria from hospitals, all kinds of health care centers (human and veterinary clinics, dentistry clinics), food processing industries, and animal farms, and the necessity of assuring certain quality of surface disinfection. In addition, this study sheds the light on the activity against the sessile form of growth, which is considered a main reason of the spread and the persistence of bacteria on medical/industrial surfaces. We assume that the tested wipes have no effect on the dispersion and adhesion of the bacterial biofilms of *S. aureus* and *P. aeruginosa* on PVC surface.

Materials and methods

Bacterial strains

The reference strains *S. aureus* ATCC 6538 and *P. aeruginosa* ATCC 15442 were used for this study. The strains stored in Microbank™ storage system (Pro-Lab Diagnostics, Richmond Hill, ON, Canada) at −80°C, were reactivated in tryptic soy broth (TSB, Oxoid, Milan, Italy) and incubated at 37°C overnight under aerobic condition. After the incubation, broth cultures were refreshed in TSB (1:10) for 2 h in water bath (37°C) with shaking (120 rpm).

Then, broth cultures were standardized in TSB by spectrophotometer (Eppendorf, Milan, Italy) to obtain optical density of $OD_{600} = 0.8$ (from 1.5×10^9 to 5.0×10^9 colony-forming unit CFU.mL^{-1}) and used separately for the experiments. The standardized broth cultures were used for biofilm study at 3 h (early attachment phase condition) and 24 h (formed biofilm).

Disinfecting wipes

The commercial disinfecting wipes "LH SALVIETTE®" were provided by Lombarda H S.r.l. (Albairate, Milan, Italy) with dimensions (20 cm × 20 cm) consisted of a mixture of biocides (Table 1). The recommended application time is 60 s. LH SALVIETTE are non-woven fabric wipes that have been soaked in a disinfectant solution containing didecyl-dimethyl ammonium chloride (DDAC) and o-phenyl phenol (OPP). This product is classified as class IIa medical device and ready-to-use (RTU) for cleaning dental and medical clinics, disinfecting clean medical devices, decontaminating medical devices prior to cleaning and sterilization, and cleaning hospital

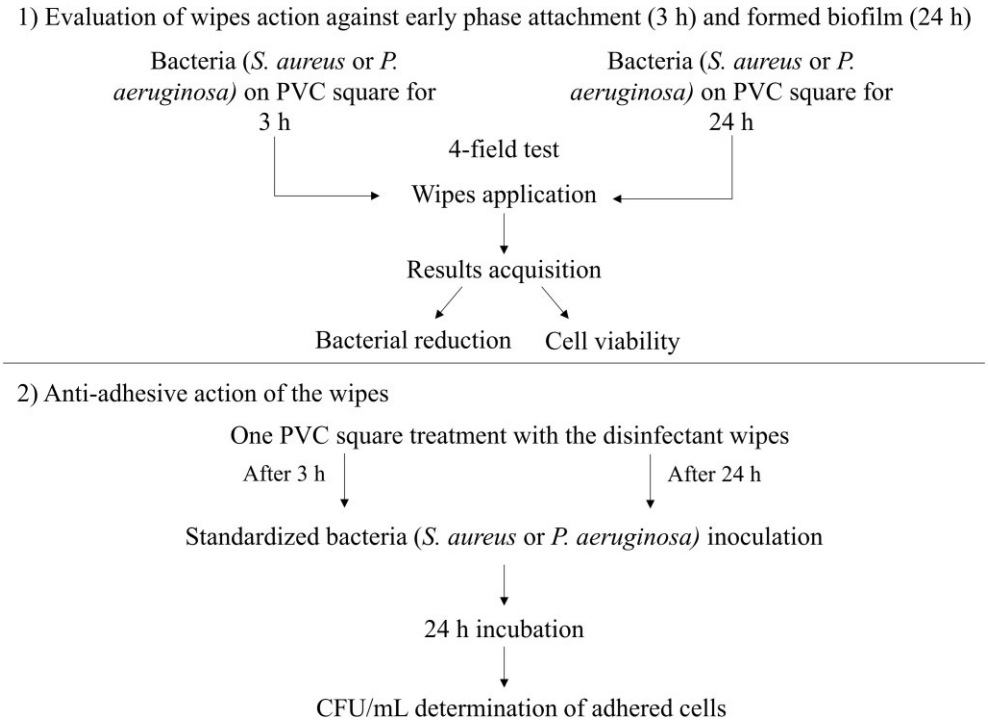

Figure 1. Study design followed in this study.

equipment and the operating1 room (hand pieces, trolleys, basins, beds, furniture, armchairs, and shelves, among others).

Evaluation of wipes action against early attachment and formed biofilms

The tests were performed according to the European Standard EN 16615:2015 (EN 16615. 2015) by using bacteria in a sessile growth phase following the study design in Fig. 1.

This is a carrier method in which disinfected surfaces are simulated by homogeneous polyvinyl chloride (PVC) panels (Fig. 2). For the experiments, all PVC panels and squares were sterilized by UV light. For analysis on PVC panels, obtained locally, four squares as test fields (T1, T2, T3, T4) each measuring 6.25 cm^2 (2.5 cm × 2.5 cm), were marked on the panel. The first square (T1) was cut out and used separately as surface for biofilm growth and placed into a sterile Petri plate (3.5 cm of diameter) and inoculated with 2 mL of each bacterial suspension (*S. aureus* or *P. aeruginosa*), in the presence of a 3 $g.L^{-1}$ bovine albumin solution (Sigma, St. Louis, MO, USA) and 3 $mL.L^{-1}$ of sheep erythrocytes simulating high organic burden condition. The first square was incubated at 37°C in aerobic condition for *S. aureus* and *P. aeruginosa* in humidity condition to avoid excessive evaporation. LH wipes effect was evaluated at two different incubation times: 3 h (early attachment phase) and 24 h (formed biofilm). After incubation times, the planktonic bacteria were removed from the T1 square by washing with PBS (Merk, KGaA, Darmstadt, Germany) and left to dry. The dried T1 square was repositioned into the PVC panel. A unitary weight weighing 2.5 kg, which provides adequate pressure on the surface, was covered with "LH SALVIETTE" wipe, and it was used for the wiping process on the PVC panel, beginning in front of test square T1 up to T4 and backward to T1 square within 2 s (Fig. 2a). In this study, three controls were used:

- ☐ Water control: it was used "17c m x 30 cm" sterile wipe provided by Lombarda H, soaked with 16 mL of sterile water (Fig. 2b)
- ☐ Sterility control: The same procedure was performed without using bacteria (Fig. 2c).
- ☐ Biofilm control: One PVC square of 6.25 cm^2 was inoculated with each standardized broth culture and used as positive control biofilm (C+) (Fig. 2d).

The effect was evaluated in terms of Log_{10} reduction and cell viability.

Log10 reduction

After wiping and waiting for contact time of 60 s, T1 square was placed in 30 mL capacity polypropylene sterile containers (sterilized glass beads included inside) with 1 mL of neutralizer (30 $g.L^{-1}$ Polysorbate 80; 3 $g.L^{-1}$ lecithin; 1 $g.L^{-1}$ L-histidine; 30 $g.L^{-1}$ saponin in Tryptone Sodium Chloride-TSC-diluent) or TSC for the controls, sonicated for 4 min and vortexed for 2 min (to release the attached bacterial cells from the surface), then serially diluted in TSC and 3 drops of 10 µL of each dilution were plated onto Tryptic Soy Agar (TSA; Oxoid, Thermo Fisher Scientific, Newport, UK). Reduction in bacterial count in T1 square, expressed as Log_{10} reduction, was calculated as the difference between the number of Log_{10} of bacteria recovered from untreated and treated samples: Log_{10} reduction = Log_{10} ($CFU_{untreated}$)—Log_{10} (CFU_{T1}). For test fields T2-T4, the bacteria were recovered from each square with cotton stick swabs. For each square, we used two swabs and one tube with a neutralizer. First, the test field was rubbed with one swab stick soaked with the neutralizer to block the action of the disinfectant wipes. This procedure was repeated twice, and the tip of the swab stick was cut off and put in the neutralizer tube. Next, the test field was rubbed with a dry swab, which was also placed in the neutralizer tube, and the tube was

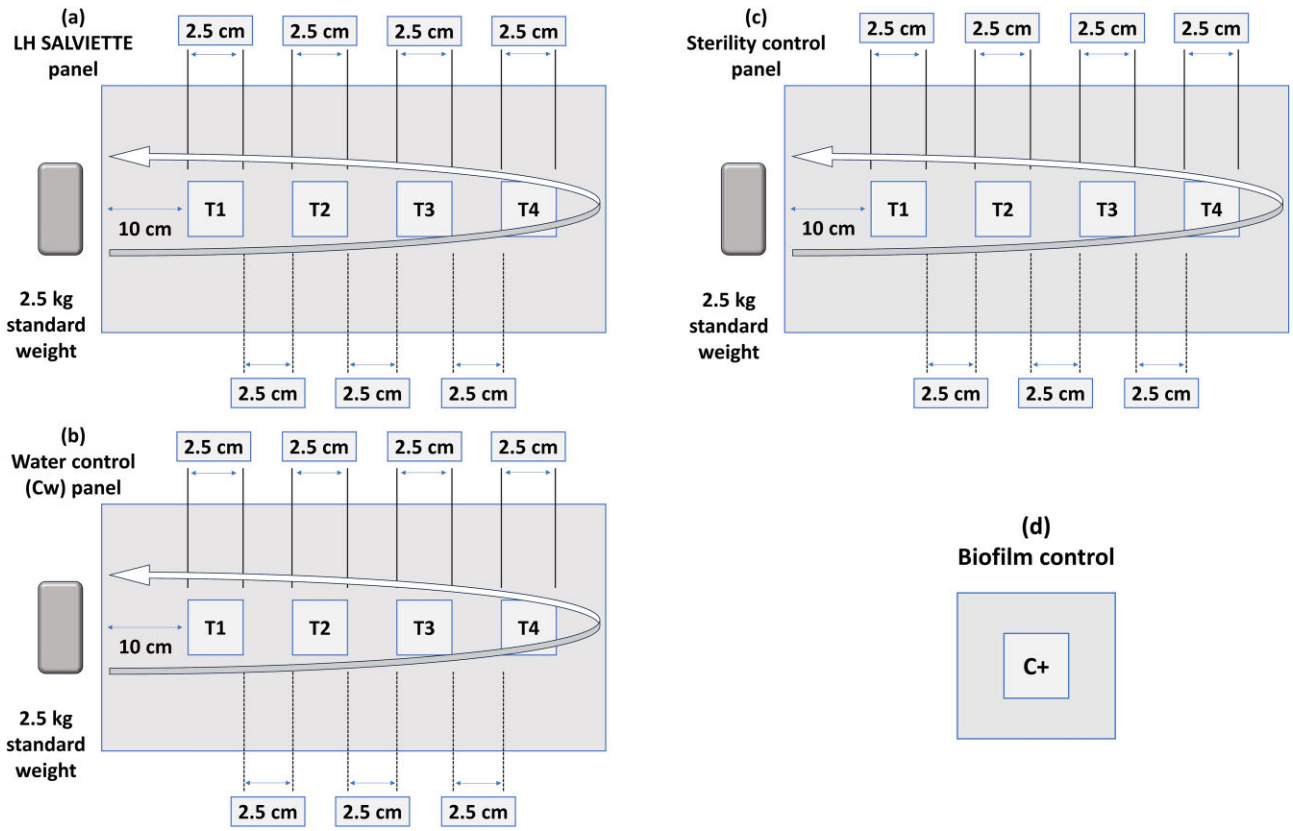

Figure 2. Schematic representation of the 4-field test PVC surface used in the study for the evaluation of wipes action: (a) LH SALVIETTE disinfecting wipes panel; (b) water control panel; (c) sterility control panel; (d) positive control biofilm square.

vortexed, then serially diluted in TSC, and 3 drops of 10 µL of each dilution were plated onto TSA and incubated at 37°C for 24 h. The bacterial load was counted and expressed in Log_{10} reduction, and the percentage of reduction of the treated samples was compared with the water control.

Cell viability assay

For the evaluation of cells viability in formed biofilms after wiping, a BacLight Live/Dead Viability Kit (Molecular Probes, Invitrogen detection technologies, USA) was used as indicated by the manufacturer. SYTO 9 stains viable cells with a green fluorescent signal, and propidium iodide stains cells with impaired membrane activity red. The images observed at fluorescent Leica 4000 DM microscopy (Leica Microsystems, Milan, Italy) were recorded at an emission wavelength of 500 nm for SYTO 9 (green fluorescence) and of 635 nm for propidium iodide (red fluorescence). Three independent experiments on PVC squares for each condition were performed and several random fields were examined.

Anti-adhesive action of the wipes

In order to evaluate the wipes capability to prevent the bacterial adhesion over time (3 h and 24 h), a PVC square (2.5 cm × 2.5 cm) was first treated with LH SALVIETTE wipe and placed into sterile Petri dishes (3.5 cm of diameter). After 3 h or 24 h from the treatment, the PVC square was covered by 2 mL of the standardized bacterial suspension (*S. aureus* or *P. aeruginosa*), in the presence of high organic load, and incubated aerobically at room temperature for 24 h. After incubation, the planktonic cells were removed from PVC square and the adhered cells were washed with sterile PBS, then the PVC square was placed in 30 mL capacity polypropylene sterile containers (sterilized glass beads included inside) with 1 mL of neutralizer (or TSC for the control), sonicated for 4 min and vortexed for 2 min, and then serially diluted in TSC, and 3 drops of 10 µL of each dilution were plated onto TSA and incubated at 37°C for 24 h as shown in Fig. 3.

Statistical analysis

Data were analyzed with Microsoft Excel 2007. All data were expressed as the mean ± standard deviation of three independent experiments in triplicate. The statistical significance of the obtained differences was evaluated using *t* student test. Values of $P < 0.05$ were considered statistically significant.

Results

Evaluation of wipes action against early attachment and formed biofilm

Log10 reduction

The antibiofilm activity of the disinfecting wipes was evaluated against *S. aureus* and *P. aeruginosa* in early attachment phase (at 3 h incubation) and the formed biofilm (at 24 h incubation) in terms of Log_{10} reduction (Fig. 4). The disinfecting wipes induced a significant ($P < 0.05$) reduction in the *S. aureus* bacterial count recovered from T1 square in respect to the biofilm control (C+) with a reduction of 1.56 Log_{10} (21.66%) after 3 h of incubation (Fig. 4a) and 1.81 Log_{10} (23.98%) in

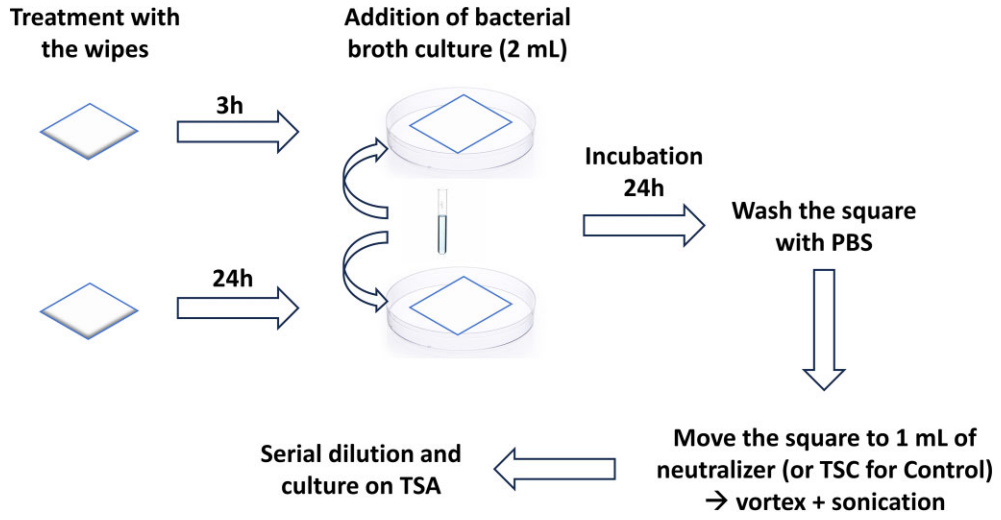

Figure 3. Schematic representation of the anti-adhesive action test of the wipes.

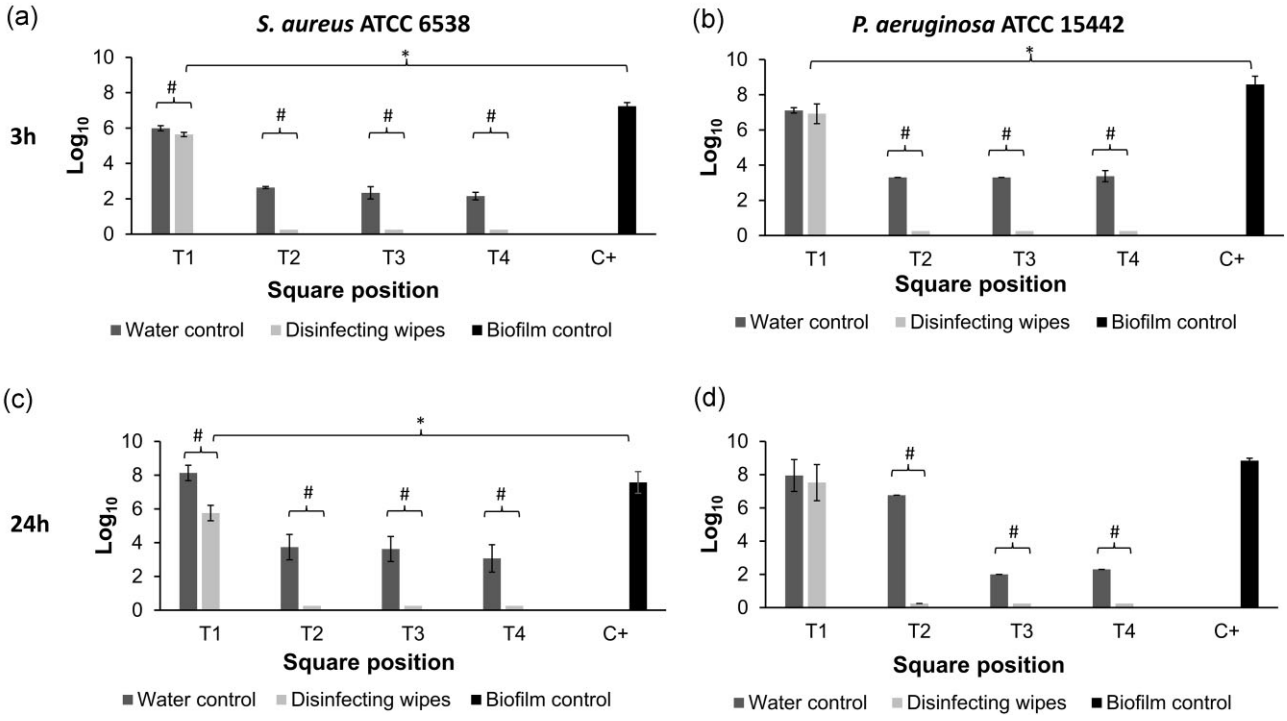

Figure 4. The disinfecting wipes activity against the 3 h (early attachment phase) and 24 h (formed biofilm) of *S. aureus* (a, c) and *P. aeruginosa* (b, d).

24 h biofilm (Fig. 4c). Similarly to *S. aureus*, the disinfecting wipes induced a reduction of 1.67 Log_{10} (19.44%) in *P. aeruginosa* after 3 h incubation (Fig. 4b) and 1.32 Log_{10} (14.96%) in the 24 h biofilm (Fig. 4d) in respect to the biofilm control (C+). Interestingly, compared to the samples treated with water, it is important to underline that there was no growth in the squares T2, T3, and T4 treated with LH SALVIETTE for both bacterial strains.

Cell viability

Live/dead images showed double activity of the disinfecting wipes on early attachment phase and formed biofilm of both *S. aureus* (Fig. 5) and *P. aeruginosa* (Fig. 6). As shown in the figures, the disinfecting wipes caused a disaggregation of the bacterial clusters and killing action compared to the untreated control.

Anti-adhesive action of the wipes

Figure 7 shows the disinfecting wipes anti-adhesive activity on PVC surface against both studied bacteria (preventive activity). Results demonstrate a significant effect of the wipes towards bacterial attachment process, especially when *S. aureus* incubated after 3 h from the PVC surface pre-treatment with the disinfecting wipes. The surface disinfection reduced the $CFU.mL^{-1}$ 99.95% in respect to the untreated surface (Fig. 7). At the same condition, a reduction of 61.09% of $CFU.mL^{-1}$ was recorded for *P. aeruginosa* in respect to the untreated surface during similar application time (3 h). After 24 h from

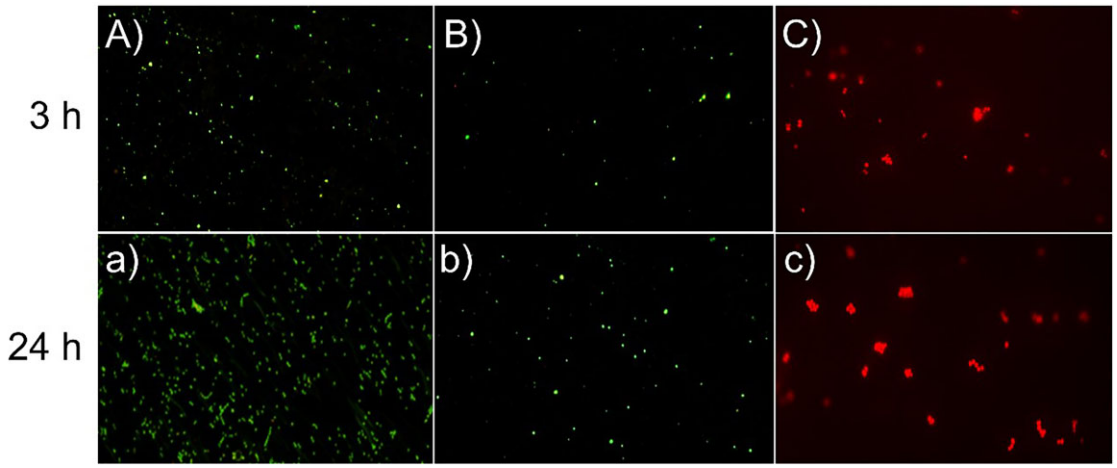

Figure 5. Representative Live/dead images of *S. aureus* early attachment at 3 h and formed biofilm at 24 h: biofilm control (A, a), T1 square of water control (B, b), T1 square of LH SALVIETTE wipes (C, c). (Original magnification, 1000X).

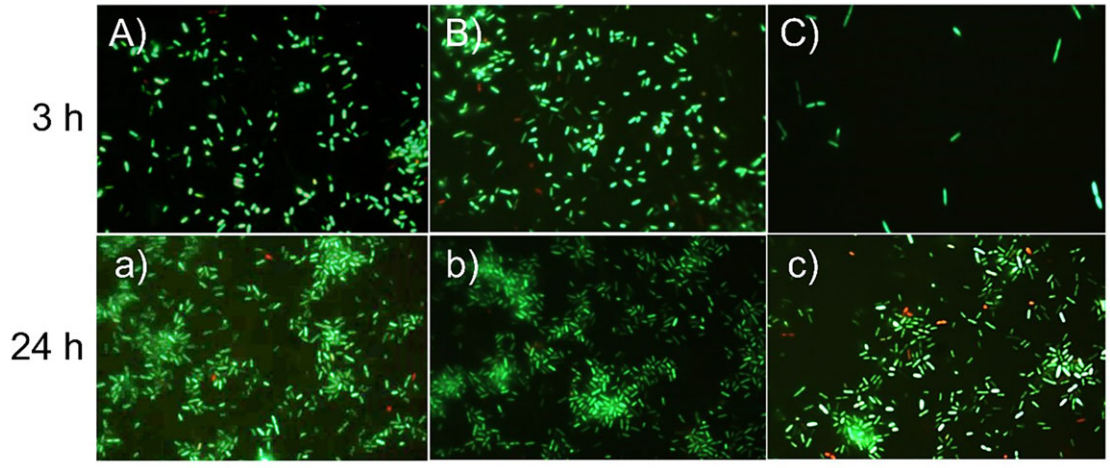

Figure 6. Representative Live/dead images of *P. aeruginosa* early attachment at 3 h and formed biofilm at 24 h: biofilm control (A, a), T1 square of water control (B, b), T1 square of LH SALVIETTE wipes (C, c). (Original magnification, 1000X).

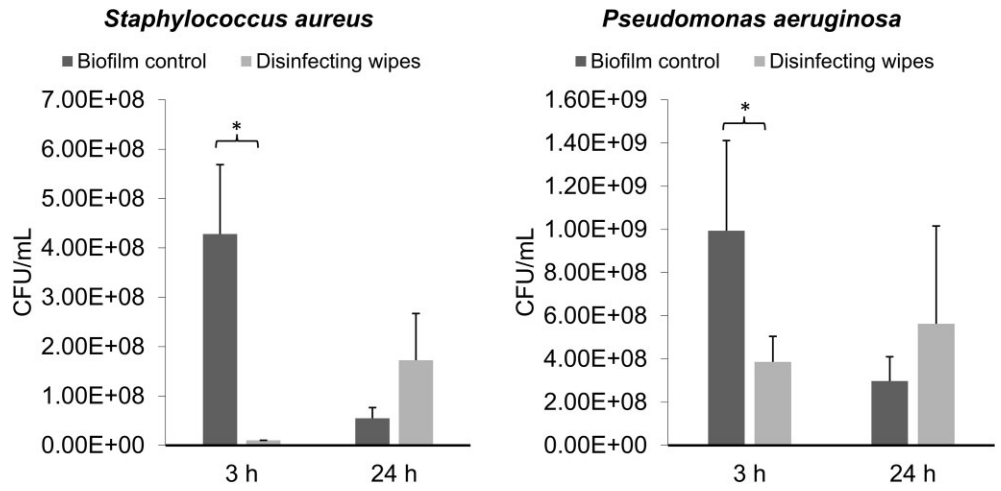

Figure 7. LH SALVIETTE anti-adhesive activity against *S. aureus* and *P. aeruginosa* after 3 h and 24 h from the pre-treatment. ∗Result statistically significant ($p < 0.05$) in respect to the control.

pre-treatment, no statistically significant difference between the treated and untreated squares was recorded for both bacteria, as shown in Fig. 7.

Discussion

The microbial colonization of the surfaces in the healthcare facilities, farms, and food industry represents a serious issue since the highly touched surfaces could be a possible carrier of several resistant bacterial cells and could increase the microbial transmission requiring an appropriate management (Brenciani et al. 2022, Grooters et al. 2024). In addition, some microbial species can survive for long time on dry surfaces and show resistance against biocides, which contribute to increased risk of infection (Costa et al. 2019).

Biofilms formed on the surfaces provide high tolerance to the embedded microorganisms against disinfectants. In the hospital settings, the biofilm formation on the reusable medical devices gives a stronger structure because of the repetitive exposure to either heat or disinfectant, which cause fixation of multiple layers of matrix over each other (Alfa 2019). Surface disinfection in health care facilities is important and involves several methods and materials in order to minimize the pathogenic colonization on these surfaces. The disinfectants can be applied directly on the surface and spread by applying mechanical action through wiping, or it is possible to use remoistened, RTU wipes (Boyce 2021). To our knowledge, this is the first work that evaluates the effect of RTU disinfectant wipes on sessile phase following the 4-field test (EN 16615:2015).

In this work, we used LH SALVIETTE wipes, RTU nonwoven fabric wipes with disinfectant mixture of DDAC and OPP following the European standard EN 16615:2015 (a European Standard that evaluates the activity of disinfectant wipes against the planktonic bacteria) with some modifications, against the bacterial sessile phase of *S. aureus* and *P. aeruginosa*.

In particular, the aim was to evaluate the anti-dispersion and anti-adhesive capabilities of these disinfectant wipes. *Staphylococcus aureus* and *P. aeruginosa* are considered important causative of HAIs, especially for their ability to remain on the surface and to form biofilm. *Staphylococcus aureus* colonizes the skin and the mucosal surfaces; moreover, the carriage of *S. aureus* is considered as risk factor for nosocomial infections. Likewise, *P. aeruginosa* is responsible for nosocomial infections, especially in the burn wounds and cystic fibrosis patients and for its antimicrobial resistance, and ability to form biofilms in hospital environments. The results of the study rejected the hypothesis under test. In particular, the disinfectant wipes were evaluated in terms of anti-dispersion and anti-biofilm proprieties on early attachment phase (3 h) and formed biofilms (24 h) of *S. aureus* and *P. aeruginosa*. The wipes reduced the bacterial count of adhered bacteria on the PVC surface demonstrating its antibiofilm activity. On the other hand, the wipes affected *P. aeruginosa* early attachment with a significant reduction compared to the positive control. There was no statistical significance when the wipes were applied on the *P. aeruginosa*-formed biofilms (24 h). Therefore, wipes are not particularly effective against already formed biofilm, but they are strongly recommended in their removal effect of microorganisms that can later form biofilms. For instance, Tyski et al. (2021) performed the 4-field test (EN 16615:2015) in evaluating the efficacy of disinfecting wipes against the planktonic phase of reference bacterial strains (*S. aureus* ATCC 6538 and *P. aeruginosa* ATCC 15442), showing a higher reduction of bacterial load in planktonic phase compared to our results related to sessile phase. These outcomes underline that the biofilm structural barrier provides increased tolerance to the sessile bacteria in comparison to the planktonic cells.

QAC and OPP efficacy were tested against nosocomial bacteria under different conditions, concentration, contact time, and method of application (Montagna et al. 2019, Ramzi et al. 2020). These compounds affect the bacterial cell at different levels. DDAC was shown to interact with the cytoplasmic membrane of *S. aureus* causing an immediate leakage of the intracellular components. In addition, DDAC induced autolysis of *S. aureus* cells and was less sensitive to temperature changes (Boyce 2023, Zhou et al. 2023). QAC showed greater efficacy against Gram-positive than Gram-negative bacteria (Song et al. 2019). Our results underline the lower efficacy against *P. aeruginosa* 24 h old-formed biofilm in respect to the 3 h old attached cells, while the disinfecting wipes affected *S. aureus* 3 h and 24 h attached cells. The limited activity of the wipes on 24 h biofilm of *P. aeruginosa* is attributable to the limited activity of QAC on *P. aeruginosa* biofilms. In fact, as previously reported, Bridier et al. visualized the action of QAC in three clinical isolates of *P. aeruginosa* biofilm, confirming a delay in the penetration leading to a diffusion limitation that can explain the tolerance of *P. aeruginosa* biofilm to this biocide (Bridier et al. 2011).

In this work, we also evaluated the cross-contamination caused by the wiping activity by wiping process on PVC panel from T1 to T4 and backword to T1 square within 2 s. Noteworthy, the data showed that there was no bacterial growth in the adjacent fields (T2–T4). It is clear that the wipes prevented the transmission of bacteria to the other test fields, which is important and crucial in surface disinfecting process. This treatment guarantees prevention from the *S. aureus* and *P. aeruginosa* biofilm dispersion in PVC surfaces.

In the second part of our work, it was tested the capability of the pretreatment with wipes to affect the adhesion of bacteria on PVC surface, during the time. The adhesion represents the first step in colonization, and, consequently, it is important to prevent this step in order to limit the bacterial persistence (Bowler et al. 2020). We tested the anti-biofilm formation action of the wipes in order to suppress the attachment step and interrupt the biofilm formation. The data showed that this positive effect is significant after 3 h of LH SALVIETTE wipes treatment. After 24 h from LH SALVIETTE wipes treatment, the bacterial adhesion was not significantly affected. The loss of efficacy at 24 h, in terms of anti-adhesive propriety, could be related to the residual organic load deposition over the time on disinfected surfaces that can negatively influence the antibacterial efficacy (Araújo et al. 2013). These data demonstrate the importance of frequent cleaning applications in healthcare facilities. Due to the difficult implementation of multiple applications of the disinfectant, researchers are testing a continuously active disinfectant that provides sustained activity on surfaces after one application (Redmond et al. 2022)).

The disinfection of surfaces in health care facilities is essential to prevent the colonization and spread of nosocomial pathogens. RTU LH wipes impregnated with disinfectants represent an important method for disinfecting surfaces. Our results showed that the wipes decreased the bacterial count after the application on PVC surface using mechanical action

against both early attachment and formed biofilms avoiding the bacterial dispersion in the PVC surfaces. The data also elaborated the importance of repeated application of the wipes to decrease the biofilm formation on surfaces.

The use of pre-impregnated disinfecting wipes represents one of the most efficacious methods for the decontamination of highly touched surfaces and non-critical medical devices in hospitals and other healthcare settings. The wiping treatment with LH SALVIETTE wipes has proven to have antimicrobial and antibiofilm activities against early attachment and formed biofilm of two relevant microorganisms, *S. aureus* and *P. aeruginosa*, avoiding the biofilm dispersion on adjacent surfaces. Moreover, the pretreatment of surface with LH SALVIETTE wipes reduced significantly the bacterial adhesion in short-time application.

In conclusion, the tested commercial LH SALVIETTE wipes could be a good and versatile solution to decrease cross-contaminations in hospital settings as well as in environments exposed to contamination by etiologically relevant pathogens. Considering the fundamental role of prevention in the One Health approach, the suggestion to use disinfecting wipes could help to control sessile microbial proliferation, avoiding potential contaminations.

Further studies could be carried out, including more pathogens, bacteria, and fungi, on different surfaces contaminated with environmental contaminants and with other pathogens simultaneously.

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Author contributions

Paola Di Fermo (Conceptualization, Formal analysis, Writing – original draft), Firas Diban (Conceptualization, Investigation, Writing – original draft), Elisabetta Ancarani (Writing – review & editing), Kelvin Yu (Writing – review & editing), Sara D'Arcangelo (Data curation), Simonetta D'Ercole (Formal analysis), Silvia Di Lodovico (Formal analysis, Funding acquisition), Mara Di Giulio (Funding acquisition, Writing – review & editing), and Luigina Cellini (Funding acquisition, Writing – review & editing).

Conflict of interest: Paola Di Fermo, Firas Diban, Elisabetta Ancarani, Kelvin Yu, Sara D'Arcangelo, Simonetta D'Ercole, Silvia Di Lodovico, Mara Di Giulio, and Luigina Cellini declare that they have no conflicts of interest relevant to the content of this manuscript. Elisabetta Ancarani and Kelvin Yu contributed to the manuscript revision since they are partners in the PhD project of Firas Diban.

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Data availability

The datasets used and/or analyzed during the current study are available from the corresponding author upon reasonable request.

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