Persistent Bactericidal Action by a Silver Disinfectant on Surfaces of Hospital Furniture

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Authors’ contributions
This work was carried out in collaboration between all authors. Author SDL managed the analyses of the study, author IB performed the statistical analysis. Author GS the first and the final draft of the manuscript. Author GF designed the study and wrote the protocol. Author PC managed the literature searches. All authors read and approved the final manuscript.

ABSTRACT

Aims: In hospitals, surfaces are often colonized by potentially pathogenic microorganisms which can remain alive for long periods of time, thus playing a major role in hospital-acquired infections. One way to overcome this drawback could be to use disinfectants with long-term action. Recent studies have shown that not only do disinfectants containing silver present an immediate effect, reducing the surface bacterial load, but that this action also appears persistent in time. This work assesses the bactericidal activity of a long-lasting disinfectant complex composed of silicon oxide, silver ions and a cationic surfactant (BACTERCLINE ENERGY BLAST) applied on the surfaces of two surgery ward rooms and left in place for different amounts of time (15 min and 72 hours).

Study Design: An experimental study.

Place and Duration of Study: The investigation was carried out in two rooms of a surgical ward at the Villa Erbosa health care facility in Bologna (Italy) over a period of seven weeks.

Methodology: The samples were taken using contact plates (diameter: 24 cm²) containing an adequate agar culture medium (Tryptic Soy Agar). After incubation at the temperature of 36±1°C for 24 and 48h, the number of colonies was counted and the statistical analysis of results was performed.

Results: The product was able to achieve a high degree of decontamination (around 90%)

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immediately after application and that, after 72 hours, decontamination remained at about 55%. The results are statistically significant.

**Conclusion:** It may be concluded that the tested product could be profitably used to decontaminate surfaces in hospital wards.

**Keywords:** Persistent silver disinfectant; surface decontamination; microbicidal effects; hospital infection prevention.

**1. INTRODUCTION**

In hospitals, surfaces are often colonized by potentially pathogenic microorganisms [1,2] which can remain on the surfaces for long periods of time [3,4], thus playing a major role in hospital-acquired infections [5,6]. Moreover, while direct transmission of nosocomial pathogens — either from one patient to the next or from the operator's hands to the patient — appears to be the most important pathway [7] to such infection. Moreover, indirect transmission — when hands come into contact with contaminated surfaces — has thus far been underestimated, a fact recently brought to light for *Clostridium difficile* [8], *Acinetobacter baumannii* [9] and MRSA [10]. Cleaning and disinfection are among the preventive measures intended to provide reassurance that patients as well as staff are not put at risk. By the proper use of disinfectants, transmission of micro-organisms from surfaces to patients (mostly via healthcare-workers' hands) can be at least partially prevented. Thus, the routine use of disinfectants to disinfect hospital microfloors and other non-critical items is controversial. Some authors believe that it is reasonable to use hospital disinfectants on non-critical patient care surfaces, patient equipment surfaces and housekeeping surfaces in patient care areas. In their critical reply, some others summarized the arguments against routine surface disinfection instead of surface cleaning without the use of biocidal substances, especially the risk of allergy and the selection for resistance, which poses a risk to humans and the environment [11].

The use of disinfectants provides a temporary reduction in the bacterial load. However, this action does not last in time because, once deposited on the surfaces, common disinfectants cannot withstand environmental aggression such as contact with water and wiping. One way to overcome this drawback could be to use disinfectants with long-term action.

In this regard, nanotechnology is a most promising field for generating new applications in medicine. A most prominent nanoproduct is nanosilver. Nanosilver particles are generally smaller than 100 nm and contain 20–15,000 silver atoms. As is the case with all nanomaterials, the principle characteristic of silver nanoparticles is their ultra small size. Ultra small particle size leads to ultra large surface area per mass where a large proportion of atoms are in immediate contact with ambiance and readily available for reaction [12]. Recent studies have shown that not only do disinfectants containing silver present an immediate effect, reducing the surface bacterial load, but that this action also appears persistent in time [13,14].

The present work seeks to assess the bactericidal activity of a long-lasting disinfectant complex composed of aqueous suspensions of nanoparticles (average diameter: 150-200 nm) of silicon oxide, silver ions in the form of silver acetate (0.005%), a cationic surfactant, benzalkonium chloride (0.6%) and 12% ethyl alcohol (BACTERCLINE ENERGY BLAST-Esoform S.p.a., Rovigo, Italy) applied on the surfaces of two surgery ward rooms and left in place for different amounts of time.
2. MATERIALS AND METHODS

2.1 Study Location

A bi-weekly investigation was carried out in two rooms of a surgical ward at the Villa Erbosa health care facility in Bologna (Italy) over a period of seven weeks. The room temperature was about 25°C and the daylight exposure was about 13 hours.

2.2 Wiping Methods

During the first 4 weeks, the cleaning contractor personnel were supplied with non-woven fabric cloths (LBK 45 g/m²) composed of wood pulp, viscose and PEP (Magris S.p.a Seriate, Bergamo, Italy) and these were used for cleaning operations. In the following 3 weeks, the manufacturer of the disinfectant supplied the contractor with 80% polyester-20% polyamide non-woven cloths (Falpi S.r.l. Trivero Ponzone, Belluno, Italy), again used to wipe down the surfaces after being moistened with tap water. The use of two different cloths is justified by the fact that not all materials are compatible with the disinfectant. For example, cotton deactivates the silver ions while, given their abrasiveness, microfiber cloths could quickly damage the silver-containing film on the surfaces treated.

2.3 Methods of Sampling and Culture

The examined surfaces were smooth, undamaged and made of plastic laminate. Samples were taken prior to decontamination, as well as 15 minutes and 72 hours after decontamination with the product being tested. After wiping down with tap water and drying, the undiluted product in the dispenser was uniformly sprayed over the surfaces to be tested and then distributed with a cloth, ensuring that a uniform film covered the entire surface. During the 72 hours period running from application of the product to the last sampling, the surfaces only underwent daily wiping with a cloth moistened simply with tap water since a detergent could have interacted with the BACTERCLINE ENERGY BLAST film, voiding its effectiveness. In both rooms, the sampling points, both before and after decontamination, were located in the following critical sites:

- three points on the table;
- two points on the chair;
- six points on the nightstand (3+3);
- three points on the surface of the trolley.

The samples were taken using contact plates (diameter: 24 cm²) containing an adequate agar culture medium (Tryptic Soy Agar) and a neutralizer (lecithin 3 g/l; polysorbate 80 30 g/l V/V; sodium thiosulfate 5 g/l; L-histidine 1 g/l; saponine 30 g/l in diluents at 1% V/V). Samplings were performed by the same operator using a Rodac-Weight (International pbi S.p.a. – Novara, Milano, Italy), which applies the same pressure — equivalent to a standard weight of 500 gr — on the contact plate. The time of application was 10 s.

2.4 Statistical Analyses

After incubation at the temperature of 36±1°C for 24 and 48h, the number of colonies was counted, the average and standard error calculated and the reduction in bacterial load evaluated as a percentage of initial counting. In addition, statistical analysis of the results
was performed using the Student test to determine significance and, finally, probability was calculated.

3. RESULTS AND DISCUSSION

The results obtained at a series of points analyzed in the two ward rooms — for the first four weeks of the investigation — and those obtained on the surfaces of the room furnishings — over the entire duration of the study — are summarized in Table 1. The average CFU/24 cm² before disinfection was 72.74±16.17 (from 1 to 1100 CFU/24 cm²) and after decontamination it was 3.88±0.90, (from 0 to 78 CFU/24 cm²) hence the immediate reduction was 94.67%. The results obtained appear to be highly significant (t₁₁₂=4.252, p<0.001). 72 hours after application of the disinfectant, the average CFU was 31.31±3.57 (from 1 to 270 CFU/24 cm²) which is still a 56.97% reduction from the initial load and this result, too, is quite significant (t₁₁₂=2.502, p=0.013).

The results of decontamination during the following three weeks are summarized in Table 2. Just 15 minutes after use of the disinfectant, the average initial CFU/24 cm² of 31.18±7.96, (from 0 to 516 CFU/24 cm²) had dropped to 3.23±0.48 CFU/24 cm², (from 0 to 30 CFU/24 cm²). Therefore, again in this case, the microbial load was markedly lower with an 89.65% reduction. This result is highly significant (t₈₄=3.505, p<0.001). Moreover, 72 hours after application of the disinfectant, the microbial load was 12.56±3.29, (from 0 to 232 CFU/24 cm² with a 59.72% reduction). The results obtained are significant (t₈₄=2.161, p=0.032).

Table 1 Statistical analysis of the data: averages and standard errors, pre- and post-disinfection, Student t value, freedom degrees (f.d.), p value and percentage reduction for the data acquired during the first four weeks

| Sampling after | Average CFU/24 cm² | t Student | f.d. | p        | Significance | R%    |
|----------------|-------------------|----------|------|----------|--------------|-------|
| 15 minutes     | prior to disinfection: 72.74±16.17 | 3.88±0.90 | 4.252 | <0.001  | Highly significant | 94.67 |
| 72 hours       | after disinfection: 31.31±3.57 | 2.502    | 112  | 0.013    | Highly significant | 56.97 |

The product tested (BACTERCLINE ENERGY BLAST) achieved high decontamination immediately after application. This is due to the fact that application creates a continuous film of product on the surfaces and that the nanoparticles of SiO₂ bond with the silver ions and cationic surfactant molecules. The positively charged nanoparticles trap the bacteria on the surface of the film. Indeed the negative charge in the bacterial outer membrane facilitates transfer of the silver ions to the microorganisms where they bond with the membrane’s sulphhydryl groups. The silver then penetrates into the cell, accumulating until a toxic threshold is reached, killing the microorganism. Such decontamination was maintained in time although it did decline somewhat, most likely because, during the 72 hours that had elapsed since application of the product, the surfaces were wiped down with cloths dampened with tap water and this rubbing may have altered the disinfectant film adhering to the surfaces.
Table 2. Statistical analysis of the data: averages and standard errors, pre- and post-
disinfection, Student t value, freedom degrees (f.d.), p value and percentage reduction
for the data acquired during the following three weeks

| Sampling after | Average CFU/24 cm² | t       | f.d. | p      | Significance   | R%    |
|----------------|--------------------|---------|------|--------|----------------|-------|
|                | Prior to disinfection | After disinfection | Student |       |                |       |
| 15 minutes     | 31.18±7.96         | 3.23±0.48 | 3.505 | 84     | <0.001         | Highly significant | 89.65% |
| 72 hours       | 31.18±7.96         | 12.56±3.29 | 2.161 | 84     | 0.032          | Significant     | 59.72% |

After ascertaining the fact that the components of the disinfectant tested do indeed have microbicidal properties, we feel that its effectiveness can above all be ascribed to the silver ions while the surfactants only act as co adjuvant. In reality, there do not appear to be any publications indicating that the product as a whole is able to reduce (as per UNI EN 13697) initial bioburden in the 1.5-5x10⁸ range for *Pseudomonas aeruginosa* ATCC 15442, *Eschericchia coli* ATCC 10536, *Staphylococcus aureus* ATCC 6538 e di *Enterococcus hirae* ATCC 10563 on the surfaces by more than a logarithm of 4. At the same concentration present in the product (0.06%) the surfactant (benzalkonium chloride) lead to a reduction of less than a logarithm of 1. Obviously, in our test field, the reduction is less evident because the initial surface bioburden was generally quite low, as would necessarily be the case in a hospital ward when routine cleaning of the surfaces is performed correctly.

The residual decontamination on the surfaces treated with the silver-containing disinfectant has proved effective against numerous pathogenic microorganisms, even after repeated environmental stress induced by washing and wiping [13]. Likewise a pilot study - performed in two wards where the surfaces contained silver and two others where this element was absent - showed that the bacterial loads were 62% and 98% lower on the surfaces with the silver vs. similar ward surfaces treated with materials not containing silver.

Moreover, some of the surfaces left untreated but located in the vicinity of those treated with the silver also showed lower bacterial loads than those found in environments with untreated surfaces. This would indicate that the decontamination with silver-containing products were not limited solely to the materials treated, but even extended to the broader environment [14], thus decreasing the likelihood of hospital-acquired infections in the entire ward.

One limitation of this study was that the use of Tryptic Soy Agar would tend to favor growth over bacteria and discourage growth of many fungi. The high temperature and short incubation time would also favor growth of fast growing, thermophilic (heat loving) organisms.

Nonetheless the aim of the research was not to identify microorganisms, but to determine the total mesophilic load which, although indirectly, does provide data on the possible presence of pathogens. The higher the total microbial load, the greater the likelihood that pathogens will be present [15].

4. CONCLUSIONS

In light of our and other results present in the literature, we can conclude that disinfectants containing silver offer very high decontamination immediately upon application and that this effect is maintained in time despite the aggression of damp wiping with tap water. Between
one application and the next this treatment appears to offer a two-fold advantage: i) economic savings on the products previously used for daily damp wiping and ii) reduced environmental pollution from residues of the chemicals in the cleansers used instead of tap water. Then the tested product could be profitably used to decontaminate surfaces in hospital wards.

Finally, allergies to silver are quite rare and thus disinfectants containing this element offer lower risk for operators and patients than other products used for surface decontamination.

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COMPETING INTERESTS

All authors declare that no competing interests exist.

REFERENCES

1. Dancer SJ. Mopping up hospital infection. J Hosp Infect. 1999;43:85-100.
2. Lemmen SW, Hafner H, Zolidann D, Stanzel S, Lutticken R. Distribution of multi-resistant Gram-negative versus Gram-positive bacteria in the hospital inanimate environment. J Hosp Infect. 2004;56:191-197.
3. Cozad A, Jones RD. Disinfection and the prevention of infectious disease. Am J Infect Control. 2003;31:243-254.
4. Hota B. Contamination, disinfection, and cross-colonization: are hospital surfaces reservoirs for nosocomial infections? Healthcare Epidemiol. 2004;39:1182-1189.
5. Rutala WA, Weber DJ. The benefits of surface disinfection. Am J Infect Control. 2004;32:226-231.
6. Noskin GA, Stosor V, Cooper I, Peterson LR. Recovery of vancomycin-resistant enterococci on fingertips and environmental surfaces. Infect Control Hosp Epidemiol. 1995;16:577-581.
7. Boyce JM, Pittet D. Guideline for Hand Hygiene in Health-Care Settings: recommendations of the Healthcare Infection Control Practices Advisory Committee and the HICPAC/SHEA/APIC/IDSA Hand Hygiene Task Force. Infect Control Hosp Epidemiol. 2002;23(12 Suppl):S3-40.
8. Wilcox MH, Fawley WN, Wigglesworth N, Parnell P, Verity P, Freeman J. Detergent versus hypochlorite cleaning and Clostridium difficile infection. J Hosp Infect. 2004;56:331.
9. Denton M, Wilcox MH, Parnell P, et al. Role of environmental cleaning in controlling an outbreak of Acinetobacter baumannii on a neurosurgical intensive care unit. J Hosp Infect. 2004;56:106-110.
10. Dealler S. Methicillin-resistant Staphylococcus aureus infections and colonizations in an intensive care unit apparently stopped by environmental factors. J Hosp Infect. 2004;58:238.
11. Dettenkofer M, Spencer RC. Importance of environmental decontamination - a critical view. J Hosp Infect. 2007;65(S2):55-57.
12. Chen X, Schluesener HJ. Nanosilver: A nanoproduct in medical application. Toxicol Lett. 2008;176:1-12.
13. Brady MJ, Lisay CM, Yurkovetskiy AV, Sawan SP. Persistent silver disinfectant for the environmental control of pathogenic bacteria. Am J Infect Control. 2003;31:208-214.
14. Taylor L, Phillips P, Hastings R. Reduction of bacterial contamination in a healthcare environment by silver antimicrobial technology. J Infect Prev. 2009;10:6-12.
15. Dancer SJ. How do we assess hospital cleaning? A proposal for microbiological standards for surface hygiene in hospitals. J Hosp Infect. 2004;56:10-5.

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