

# Surface disinfection: evaluation of the efficacy of a nebulization system spraying hydrogen peroxide

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## Key words

Surface disinfection • Hydrogen peroxide • Nebulization

## Summary

**Introduction.** The study assessed the efficacy of a system of nebulization of a hydrogen peroxide-based solution for surface disinfection.

**Methods.** Different concentrations (1, 2 and 4 ml/m<sup>3</sup>) of the same disinfectant solution (active principle: hydrogen peroxide) were nebulized inside a 50 m<sup>3</sup> experimental environment. Sampling was carried out on both horizontal and vertical surfaces, and the total bacterial load at 37 °C was determined by means of direct contact with Rodac plates. The disinfection efficacy of the system was evaluated by comparing the total bacterial load measured on the surfaces before and after treatment. Statistical analysis was performed by means of Stata/SE9® software.

**Results.** The percentage reduction in the mean bacterial load on horizontal surfaces as a result of treatment at concentrations of 1, 2 and 4 ml/m<sup>3</sup> proved to be 54.9%, 70.9% and 86.9%, respectively. With regard to vertical surfaces, the percentage reduction was 100% in all experimental conditions.

**Discussion and conclusions.** The system tested proved to be efficacious in disinfecting surfaces inside environments of 50 m<sup>3</sup> in volume. It could therefore be used to disinfect surfaces in hospital and community settings. In healthcare facilities, disinfection by means of nebulization systems could help to reduce the risk of spreading nosocomial infections.

## Introduction

In hospital and community settings, environmental surfaces can contribute to the spread of cross-infections, in that they constitute a possible transitory site for the accumulation of microorganisms, which may be deposited on them through contact with the hands of healthcare personnel and patients or with infected instruments and materials [1].

With regard to the efficacy of the various procedures of disinfection and sanitation carried out on surfaces in healthcare environments, the scientific literature is still scant and studies have sometimes yielded conflicting results. There is therefore no clear indication as to which method of disinfection is most efficacious in reducing the rate of hospital infections. In choosing an appropriate disinfection procedure, two essential features must be taken into account; the disinfectant used must be effective in reducing pathogenic microorganisms (broad spectrum of action) in various environmental conditions, and it must not produce by-products that are harmful to human health or have a corrosive effect on surfaces. Moreover, our knowledge of the role and efficacy of sanitation procedures in combating the ever-increasing spread of resistant pathogens in hospital settings is, as yet, inadequate [2-4].

Among the various disinfectants that satisfy most of the above-mentioned requisites are hydrogen peroxide and its compounds. These yield very good results in terms of dis-

infection efficacy and seem able to replace currently used substances that are more problematic from a toxicological standpoint, such as chlorine and its derivatives [5]. In particular, a study by Klapes et al. demonstrated the possibility of utilizing vaporized hydrogen peroxide (VPHP) to decontaminate surfaces [6].

Theilen et al. conducted tests on various commercially available disinfectants in aerosol form, and also assessed their efficacy against bacterial spores; the best results were obtained with hydrogen peroxide, peracetic acid and formaldehyde [7]. Other studies about the efficacy of hydrogen peroxide as a disinfectant were carried out through verifying hydrogen peroxide effects against *Clostridium botulinum* spores and mycobacteria [8-10]. Bacteriological monitoring of surfaces before and after treatment with a few disinfectants was carried out by Dharan et al., the best result being obtained through the use of oxygen-based and ammonium quaternary-based compounds [4].

The efficacy of disinfection needs to be monitored through a program of constant surveillance based on reports of hospital infections and, if need be, through laboratory tests carried out on bacteriological samples [4, 11, 12]. The aim of the present experimental study was to assess the disinfection capability of hydrogen peroxide in aerosol form on environmental surfaces by comparing the total bacterial load on surfaces before and after treatment by means of a nebulizer.

## Methods

The apparatus tested is a nebulizer (Nocospray® – Saluskey®) which emits an aerosol of hydrogen peroxide solution (Nocolyse®) in order to disinfect surfaces within an environment. The solution is sprayed up to a distance of several meters from the device at a velocity of about 80 m/s, and spreads throughout the environment as a result of the Venturi effect. The device is able to nebulize the non-toxic, biodegradable disinfectant solution in 0.5  $\mu\text{m}$  particles, which do not remain in the environment after treatment. The disinfectant contains hydrogen peroxide as its principal component, other ingredients being: a catalyst, biosurfactants and excipients. The device is equipped with a solution output regulator, which can be set according to the volume of the environment to be treated. As the nebulizer has an automatic on/off switch, the operator does not necessarily have to remain in close proximity to the apparatus while it is working.

The volume of the experimental environment subjected to disinfection was about 50 m<sup>3</sup>. The study was delivered in a laboratory used usually for medical and biological research. The surfaces of the laboratory were tested at the end of a working day. The surfaces, both horizontal than vertical, were not cleaned or sanitized. Five horizontal and three vertical, smooth, non-porous surfaces were tested each for five times. Before and after treatment, the total bacterial load on each surface was measured through direct contact by means of Rodac plates (24 cm<sup>2</sup>) containing, irradiated Tryptic Soy Agar

(TSA) culture medium. Sampling was carried out immediately before and 30 minutes after nebulization at a height of about 90 cm; the bacterial count was taken after 48 hours of incubation at 37°C.

A total of 120 samples were taken at different disinfectant concentrations (A, B, C), as reported below:

- A) 1 ml/m<sup>3</sup> (disinfectant consumption 50 ml; treatment duration 3 min);
- B) 2 ml/m<sup>3</sup> (disinfectant consumption 100 ml; treatment duration 6 min);
- C) 4 ml/m<sup>3</sup> (disinfectant consumption 200 ml; treatment duration 12 min).

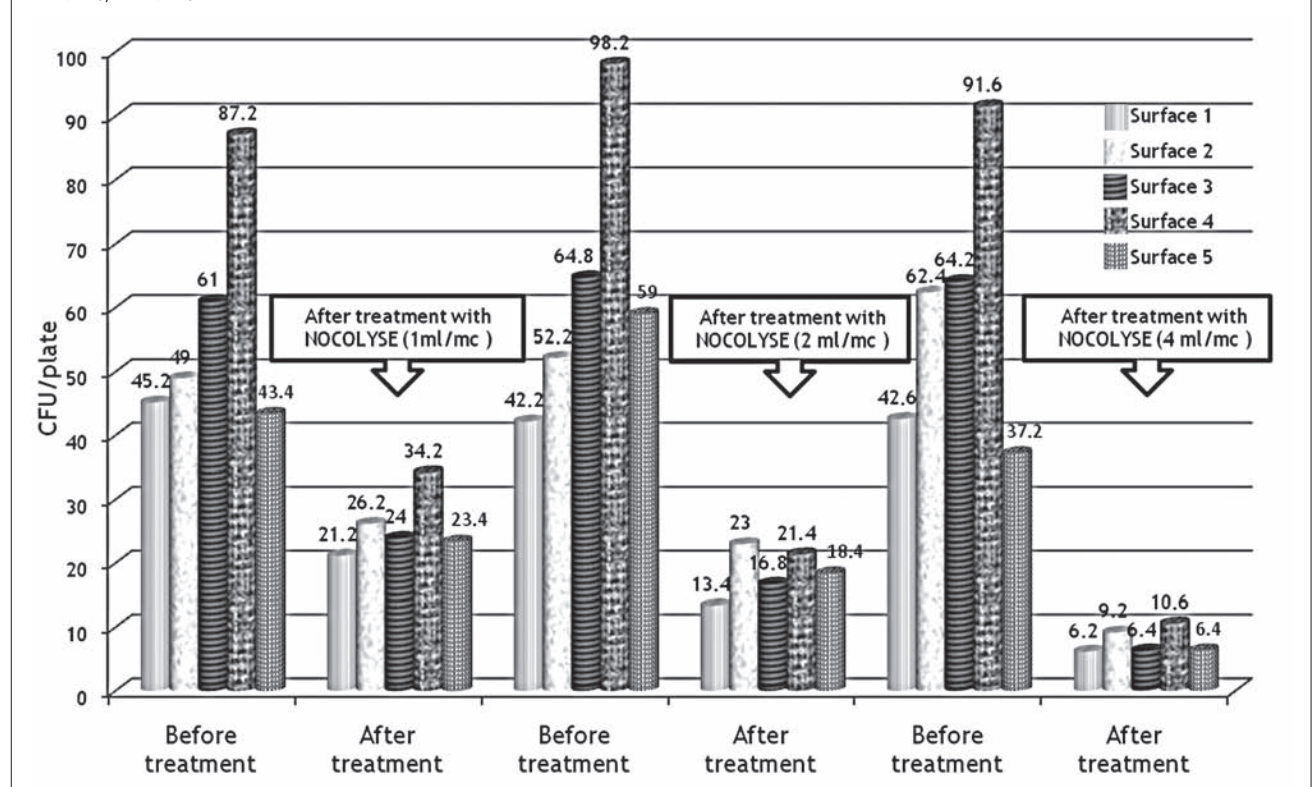
Statistical analysis was carried out by means of Stata/SE9® software.

## Results

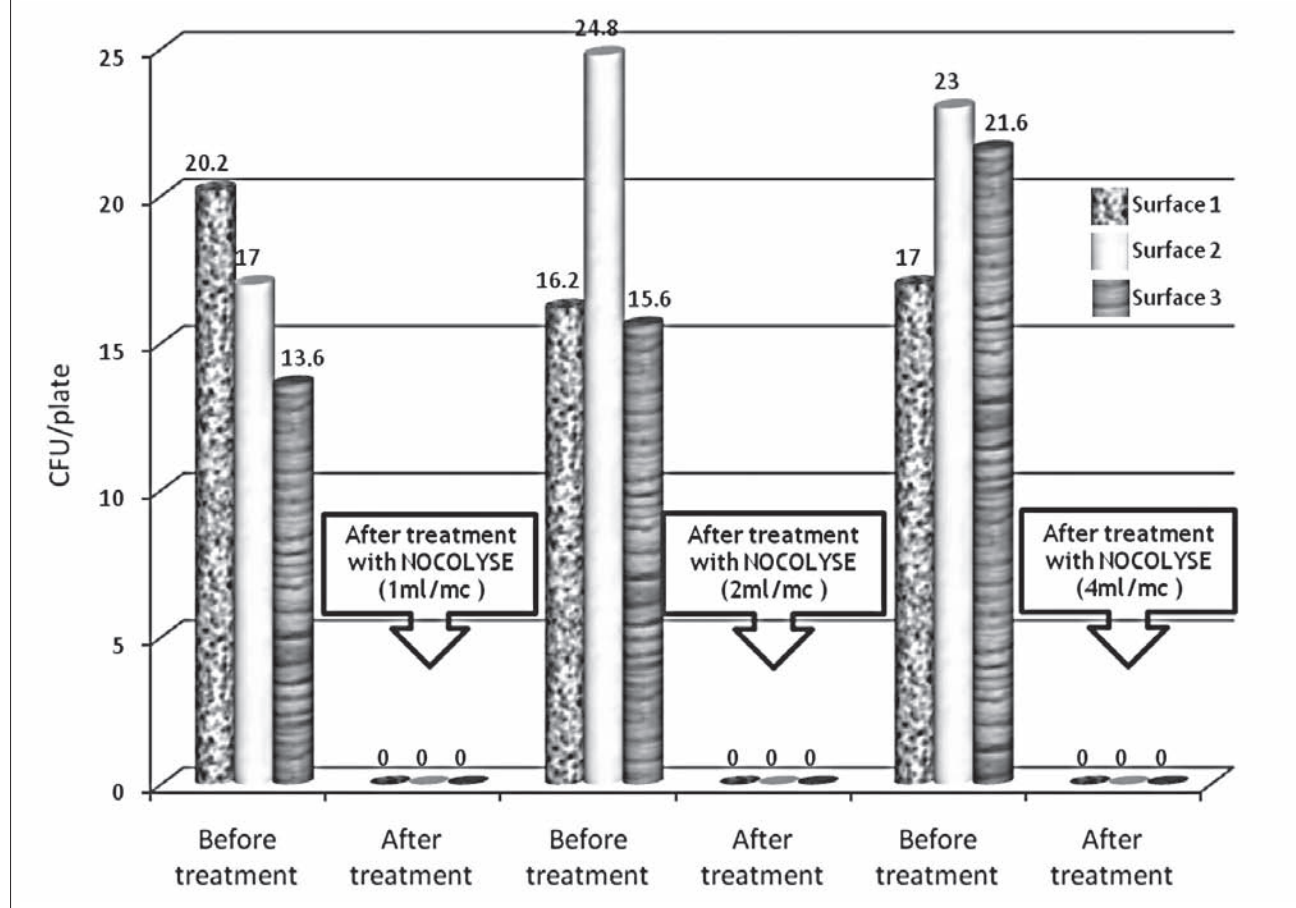
### HORIZONTAL SURFACES

The mean total bacterial load before treatment at a disinfectant concentration of 1 ml/m<sup>3</sup> was  $57.2 \pm 43.8$  CFU/plate (maximum value =  $87.2 \pm 71.0$  CFU/plate; minimum value =  $43.4 \pm 54.4$  CFU/plate). After treatment at this concentration, the mean total bacterial load on the exposed surfaces proved to be  $25.8 \pm 12.4$  CFU/plate (maximum =  $34.2 \pm 14.8$  CFU/plate; minimum =  $21.2 \pm 5.4$  CFU/plate). The mean percentage reduction in the surface bacterial load at the concentration

Fig. 1. Total bacterial load mean on the horizontal surfaces before and after treatment with Nocolyse at different concentration (1 ml/mc, 2 ml/mc, 4 ml/mc).



**Fig. 2.** Total bacterial load mean on the vertical surfaces before and after treatment with Nocolyse at different concentration (1 ml/mc, 2 ml/mc, 4 ml/mc).



of 1 ml/m<sup>3</sup> was therefore 54.9% (maximum value = 60.8%; minimum value = 46.1%).

Next, the effect of disinfection at a concentration of 2 ml/m<sup>3</sup> was evaluated. Before treatment, the mean total bacterial load was 64.0 ± 66.1 CFU/plate (maximum value = 98.2 ± 118.6 CFU/plate; minimum value = 42.2 ± 15.6 CFU/plate). After treatment at this concentration, the mean total bacterial load on the exposed surfaces proved to be 18.6 ± 9.5 CFU/plate (maximum = 23.0 ± 13.3 CFU/plate; minimum = 13.4 ± 4.2 CFU/plate). The mean percentage reduction in the surface bacterial load was therefore 70.9% (maximum = 78.2%; minimum = 56.3%).

Finally, before treatment with the disinfectant at the concentration of 4 ml/m<sup>3</sup>, a mean total bacterial load of 59.6 ± 39.9 CFU/plate (maximum = 91.6 ± 51.1 CFU/plate; minimum = 37.2 ± 38.0 CFU/plate) was recorded. After treatment at this concentration, the mean total bacterial load on the exposed surfaces proved to be 7.8 ± 8.3 CFU/plate (maximum = 10.6 ± 10.0 CFU/plate; minimum = 6.2 ± 5.2 CFU/plate). The mean percentage reduction in the surface bacterial load after treatment at the concentration of 4 ml/m<sup>3</sup> therefore proved to be 86.9% (maximum = 90.0%; minimum = 82.8%).

In the Figure 1 is reported the total bacterial load mean on the horizontal surfaces before and after treatment

with Nocolyse at different concentration (1 ml/mc, 2 ml/mc, 4 ml/mc).

#### VERTICAL SURFACES

The mean total bacterial load before treatment at a disinfectant concentration of 1 ml/m<sup>3</sup> was 16.9 ± 8.5 CFU/plate (maximum value = 20.2 ± 11.6 CFU/plate; minimum value = 13.6 ± 4.8 CFU/plate). After treatment at this concentration, the mean total bacterial load on the exposed surfaces proved to be 0 CFU/plate.

Next, the effect of disinfection at a concentration of 2 ml/m<sup>3</sup> was evaluated. Before treatment, the mean total bacterial load was 18.9 ± 10.0 CFU/plate (maximum value = 24.8 ± 11.3 CFU/plate; minimum value = 15.6 ± 11.1 CFU/plate). After treatment at this concentration, the mean total bacterial load on the exposed surfaces proved to be 0 CFU/plate.

Finally, before treatment with the disinfectant at the concentration of 4 ml/m<sup>3</sup>, a mean total bacterial load of 20.5 ± 7.8 CFU/plate (maximum = 23.0 ± 3.5 CFU/plate; minimum = 17.0 ± 8.6 CFU/plate) was recorded. After treatment at this concentration, the mean total bacterial load on the exposed surfaces proved to be 0 CFU/plate. The mean percentage reduction in the surface bacterial load after treatment at all three disinfectant concentrations used was 100%.

In the Figure 2 is reported the total bacterial load mean on the vertical surfaces before and after treatment with Nocolyse at different concentration (1 ml/mc, 2 ml/mc, 4 ml/mc).

## Discussion and conclusions

The results yielded by this study revealed the efficacy of the Nocospray® – Saluskey® system of surface disinfection in the environment of about 50 m<sup>3</sup> tested. A greater reduction in the microbial load on the vertical surfaces was observed. However, it should be borne in mind that the initial level of bacterial contamination of these surfaces was lower than that of the horizontal surfaces. Overall, the high percentage reductions in the total bacterial load recorded after treatment with this disinfectant suggest that it could be used as an

auxiliary means of disinfection in hospital environments.

On the other hand, a limitation to the use of the apparatus tested is imposed by the fact that disinfection can only be carried out in unoccupied environments; this means that the device cannot readily be used in certain environments, such as intensive care units and some protected wards, which are likely to be constantly occupied. A further drawback to this device is that it is somewhat noisy.

In conclusion, given the efficacy of hydrogen peroxide in aerosol form as a surface disinfectant, the next step will be to clarify its role as a possible means of controlling hospital infections, bearing in mind the fact that the disinfection of environmental services must always be accompanied by the implementation of correct behavioral procedures (such as proper hand hygiene) on the part of both staff and patients, without which the efficacy of disinfection would be nullified.

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