ASSESSMENT OF THE EFFECT OF AEROSOL DISINFECTION OF NANOSILVER AND HYDROPEROXIDE AG$^+$ AT CAN THO UNIVERSITY OF MEDICINE AND PHARMACY

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ABSTRACT

Background: One of the crucial methods for lowering hospital infections is air disinfection. Because the airborne contagion environment is difficult to control, the development of effective air disinfectants is critical to maintaining a safe air environment and preventing respiratory infections. At the Hospital of Can Tho University of Medicine and Pharmacy, nanosilver and hydroperoxide Ag$^+$ are two different kinds of airborne infection control, so an analytical cross-sectional study was conducted with objectives: to compare the disinfection efficacy of two solutions, nanosilver and 5% Hydroperoxide - 0.005% Ag$^+$, and to assess the efficacy of the silver nano solution and 5% Hydroperoxide - 0.005% Ag$^+$ solution in disinfecting hospital air. Materials and methods: This study was conducted on 156 air microbiological samples in 13 wards at the Can Tho University of Medicine and Pharmacy Hospital using Koch's dust deposition method based on the principle of allowing dust to settle on the surface of nutrient agar boxes for a certain period. Data analysis using SPSS 26.0 software. Results: After disinfecting the rooms with nanosilver, the air quality was good in 69.23% of cases and pretty in 30.07%. With 5% Hydroperoxide - 0.005% Ag$^+$, the number of rooms with good air quality was 15.39%, with pretty air was 69.23, and with poor air was 15.39%. Nanosilver can eradicate the fungus from Sabouraud Agar medium, and the density after disinfection was lower than before; the difference was not statistically significant (p>0.05). Moreover, on Macconkey Agar and Blood Agar medium, bacterial density after disinfecting by nanosilver was decreased; the difference was statistically significant (p < 0.05). Nanosilver reduced the bacterial density in 4 types of rooms better than 5% Hydroperoxide - 0.005% Ag$^+$ solution. However, the difference was not statistically significant (p > 0.05). Conclusions: Nanosilver can be used to disinfect the hospital air as an alternative to 5% Hydroperoxide - 0.005% Ag$^+$. Nanosilver disinfects well on gram-negative and gram-positive bacteria. Additional methods for improving the antifungal impact of nanosilver are required.
Keywords: Aerosol disinfection, nanosilver, hydroperoxide Ag$^+$

I. INTRODUCTION

Bacteria may exist in many environments, but the air is the most harmful since it is easy to spread and difficult to manage. Bacteria can survive in an air environment for several hours or even months, such as the *Tuberculosis bacterium, Staphylococcus aureus*, which will increase the risk of infection and disease. Silver nanomaterial has been one of the biggest nanotechnology successes since 1980 [1]. It can be applied in some medical fields, such as histopathological changes of organs (lungs, liver, kidney, and brain) using Agcoat nanosilver or biomedical applications on infection and inflammation [2],[3]. Its antibacterial ability has been proved by some research, especially in aerosol and surface disinfection [4],[5]. Another

advantage of nanosilver is that it is a broad-spectrum antibacterial agent that is non-toxic to humans and animals at 100 ppm or less [6]. Hydrogen peroxide Ag^+ (H_2O_2), a solution composed of two hydrogen atoms, two oxygen atoms, and Ag^+, is usually used at the Can Tho University of Medicine and Pharmacy because it eliminates bacteria for a long time [7]. However, studies on the sterilizing quality of nanosilver in the air of Vietnam hospitals are very limited. So, we decided to conduct the research entitled entitled "Assessment of the effect of aerosol disinfection of nanosilver and hydroperoxide Ag^+ at Can Tho University of Medicine and Pharmacy" with the following goals: (1) Evaluation of the air disinfection standards of 5% Hydroperoxide - 0.005% Ag^+ and silver nano at the Hospital of Can Tho University of Medicine and Pharmacy. (2) Compare the disinfection efficiency of the two solutions: nanosilver and 5% Hydroperoxide - 0.005% Ag^+.

II. SUBJECTS AND METHODS

2.1. Research subjects

Air environment in 13 wards' rooms of Can Tho University of Medicine and Pharmacy Hospital.

Standard for selection: The room is not clean, has no patients, and is used frequently by medical staff and patients.

Standard for elimination: The room where the patient is being treated, medical staff room, or without the consent of the department.

$$N = \frac{Z_{1-\alpha/2}^2 \, X \, p(1-p)}{d^2}$$

N: Minimum sample size; α: Reliability (α = 0.05); Z: Confidence coefficient (Z = 1.96); d: Estimated error (d = 0.05); p: The antibacterial effect of Nanosilver is based on research by I-Jen Wang (93%) [8], so p = 0.93. Calculate the sample size n = 100 and the allowed error of 10%, equivalent to 10 samples. We collected 156 samples.

Sampling method: Convenient

Statistic method: Paired-Samples T-Test

Research content: Quantitative research sample:

By random sampling: all the individuals in the population have the same chance (same probability) to be selected; we have discrete random variables:

Independent variables: The department of the hospital of Can Tho University of Medicine and Pharmacy has not been disinfected.

Dependent variables: Density of bacteria, types of bacteria distributed in the air environment in departments of Can Tho University of Medicine and Pharmacy hospital.

The results were evaluated based on the criteria of air quality to comply with the WHO 2012, V. Omelanski standard for clean rooms [9].

Qualitative variables

Table 1. Qualitative variables

Variable definition	Classify
Sampled room temperature	Hierarchical variable
Room type sampled	Categorical variable
Area of sampling room	Categorical variable

2.2. Research Methods

(1) Collecting samples by Koch's dust deposition method is based on the principle of allowing dust to settle on the surface of nutrient agar boxes for a certain period. Each room will be tested with five clusters, one in the middle and four in the four corners. In each cluster put 3 mediums of nutritional agar. The three types of nutritional agar: One medium of blood agar, one medium of Macconkey agar and one medium of Sabouraud agar for fungi. All mediums will be opened one after another after being fully placed at 5 positions in the operating room and left in the room for 10 minutes; then the lid is closed in turn, and the medium that opens the lid first will be covered first.

(2) Incubate the agar boxes under suitable conditions.

(3) Count the number of bacterial colonies on each agar box.

(4) Evaluate the effectiveness of air disinfection according to the standards of V. Omelanski [9]:

$X < 312$ CFU/m^3 (<5 colonies/3 agar mediums) means a good result.

312 CFU/m$^3 < X < 1250$ CFU/m^3 (5–20 colonies/3 agar mediums) means pretty.

1250 CFU/m$^3 < X < 1562$ CFU/m^3 (20-25 colonies/3 agar mediums) is evaluated as average result.

$X > 1563$ CFU/m^3 (>25 colonies/3 agar mediums) is evaluated as a poor result.

The rooms of good level are of standard quality based on V. Omelanski [9].

The rooms of pretty, average, and poor levels are substandard.

(5) Identification of bacteria distributed in the research sample.

Data processing: SPSS 26.0 software.

III. RESULTS

3.1. General characteristics of the study sample

Our study included 156 samples, with 84 samples isolated at 22°C (accounting for 53.85%) and 72 samples isolated at 27°C (accounting for 46.15%). As the record, 48 samples were isolated in the surgery rooms (accounting for 30.77%), 24 samples were isolated in the clinic (accounting for 15.38%), and 36 samples were isolated in the inpatient rooms (accounting for 23.08%), and 48 samples (30.77%) were isolated in the minor surgery rooms. Regarding the area of the inoculation rooms, the number of samples in the 16m^2 room was 60 (accounting for the highest rate of 38.46%), the number of samples in the 20m^2 and 36m^2 rooms were both 24 (both accounting for 23.08%), the number of samples inoculated in room 32m^2 was 12 (accounting for the lowest rate of 7.7%), the remaining 36 samples were inoculated in rooms with other area (23.08%).

3.2. Air sterilization efficiency of nanosilver and 5% Hydroperoxide-0.005% Ag$^+$
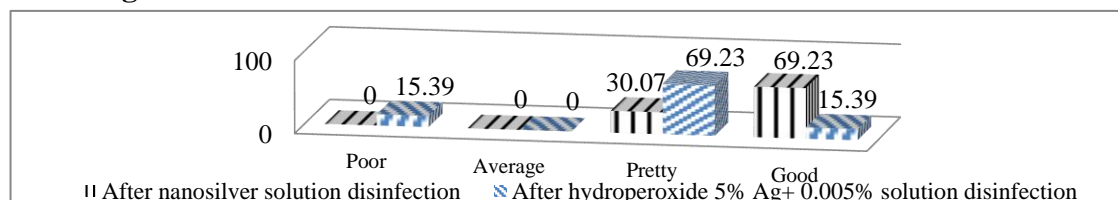

Figure 1. After disinfection, air quality of the wards with 5% Ag$^+$ 0.005% hydroperoxide and nanosilver solution.

After disinfecting the departments with Nanosilver solution, rooms with good air quality accounted for 69.23%, while the rooms with poor air quality accounted for 30.07%. After disinfection with 5% Hydroperoxide - 0.005% Ag^+, rooms with good air quality accounted for 15.39%, pretty air accounted for 69.23%, and poor air accounted for 15.39%.

Table 2. Demonstrates the variation in density of bacteria and fungus on different types of agar medium before and after sterilization with silver nano solution and Hydroperoxide 5% - Ag^+ 0.005% solution.

Agar	Nanosilver solution				Hydroperoxide 5% - Ag^+ 0.005% solution			
	Before disinfection (CFU/m^3)	After disinfection (CFU/m^3)	t	p	Before disinfection (CFU/m^3)	After disinfection (CFU/m^3)	t	p
Macconkey Agar	7.8 ± 11.3	0	1	<0.05	38.4 ± 233.7	24.7 ± 89	2.88	>0.05
Sabouraud Agar	15.7 ± 192	7.8 ± 14.1	2.5	>0.05	35.9 ± 36.5	12.3 ± 14.4	2.88	<0.05
Blood Agar	176.1 ± 143.2	41.5 ± 42.9	3.69	<0.05	286 ± 272.8	134.6 ± 102.6	1.09	<0.05
General	215.4 ± 171.5	53.2 ± 47	3.6	<0.05	421.3 ± 397.4	185.1 ± 137.3	2.96	<0.05

In summary, both techniques after sterilization are qualified according to the V. Omelanski method ($< 312 CFU/m^3$); the difference in bacterial density and fungus after disinfection compared to before sterilization is statistically significant ($p<0.05$). The difference in bacterial density before and after disinfection with nanosilver revealed that the average bacterial density on Macconkey Agar Blood Agar media dropped, and there was a statistically significant difference ($p<0.05$). Still, on SA, there was not a statistically significant. The average bacterial density on Macconkey Agar medium was eligible after disinfection with 5% HP-0.005% Ag^+; however, the difference was not statistically significant between before and after disinfection. Particularly, the average density of bacteria and fungi in BA and Sabouraud Agar medium decreased with statistically significant ($p < 0.05$).

Table 3. Change in the density of bacteria and fungi before and after disinfection in different types of rooms with silver nano-solution and Hydroperoxide 5%- Ag^+ 0.005%

Types of rooms	Nanosilver solution				Hydroperoxide 5% - Ag^+ 0.005% solution			
	Before disinfection (CFU/m^3)	After disinfection (CFU/m^3)	t	p	Before disinfection (CFU/m^3)	After disinfection (CFU/m^3)	t	p
Surgery	110.1 ± 73.8	39.3 ± 20.3	1.5	>0.05	196.6 ± 130	110.1 ± 105.1	4.1	<0.05
Clinic	228.1 ± 144.6	15.7 ± 0	2.07	>0.05	669.9 ± 700.6	283.1 ± 66.7	0.93	>0.05
Minor sugery	157.3 ± 36.3	58.9 ± 29.8	3.47	<0.05	522.9 ± 542	212.3 ± 179.1	1.7	>0.05
Inpatient room	Nanosilver solution				Hydroperoxide 5% - Ag^+ 0.005% solution			
	424.7 ± 247.2	89.1 ± 86.6	2.4	>0.05	398.4 ± 190.3	183.5 ± 154.4	7.3	<0.05

After disinfecting with nanosilver solution and 5% Hydroperoxide - 0.005% Ag+ solution, both methods generally meet the standards according to V. Omelanski method (<312 CFU/m3). The change in bacterial density before and after disinfection with Nanosilver recorded the largest decrease in the average bacterial density in the surgery room, and there was a statistically significant difference between before and after disinfection (p<0.05). The difference was not statistically significant in surgery, clinic, and inpatient rooms. Similarly, for disinfection with 5% Hydroperoxide - 0.005% Ag+ solution, a clear change was observed between the period before and after disinfection in the surgery room and inpatient room (p<0.05). In other rooms, the difference in the average density of bacteria and fungi before and after disinfecting with 5% Hydroperoxide - 0.005% Ag+ has no statistical significance (p<0.05).

Table 4. Comparison of the disinfection efficiency of two solutions of silver nano and 5% Hydroperoxide - 0.005% Ag$^+$ on aerosol disinfection.

Types of rooms	After disinfection by 5% Hydroperoxide - 0.005% Ag$^+$ (CFU/ m^3)	After disinfection by nanosilver (CFU/m^3)	t	p
Surgery	110.1 ± 105.1	39.3 ± 20.3	1.55	p>0.05
Clinic	283.1 ± 66.7	15.7 ± 0	0.4	p>0.05
Minor sugery	212.3 ± 179.1	58.9 ± 29.8	1.5	p>0.05
Inpatient room	183.5 ± 154.4	89.1 ± 86.6	0.79	p>0.05

The difference was not statistically significant between the two solutions of silver nano and 5% Hydroperoxide - 0.005% Ag$^+$ (p > 0.05).

IV. DISCUSSION

Through the survey of 156 samples, we obtained some results on the sterilization efficiency of hospital wards. The proportion of rooms with good air quality after disinfection using nanosilver solution is 69.23%, higher than disinfection with 5% Hydroperoxide - 0.005% Ag$^+$ solution (i.e., 15.39%). The number of rooms with pretty air quality using nanosilver solution and 5% Hydroperoxide - 0.005% Ag$^+$ solution was 30.07% and 69.23%, respectively. Both types of solutions give an average quality rate of 0%. Whereas the silver nano solution was 0% and the 5% Hydroperoxide - 0.005% Ag$^+$ solution was 15.39% for poor room air quality. This demonstrates that after disinfection with 5% Hydroperoxide - 0.005% Ag$^+$ solution, there is still a percentage of rooms with poor air quality. The fraction of rooms with good air quality using 5% Hydroperoxide - 0.005% Ag$^+$ solution is still low. Most rooms with good air quality results were discovered for nanosilver solution; however, there was still a considerable proportion of rooms with pretty air quality.

We discovered that both methods satisfied the requirements set by V. Omelanski after isolating three types of agars, Sabouraud Agar, MacConkey Agar, and Blood Agar, for disinfection with a nanosilver solution. The average bacterial density in the surgery room before disinfection with nanosilver solution was 110.1 ± 73.8 CFU/m^3. After disinfection, the average bacterial density was 39.3 ± 20.3 CFU/m^3; this difference was not statistically significant (p > 0.05). The average density in the clinic before disinfection was 228.1 ± 144.6 CFU/m^3, and after disinfection was 15.7 ± 0 CFU/m^3. This difference was not statistically significant (p > 0.05). These statistics show that inpatient rooms have the

highest density of bacteria. Thus, inpatient rooms air should be disinfected carefully to ensure good quality in the hospital environment.

When nanosilver was used in the surgery room, the average bacterial density before disinfection was 157.3 ± 36.3 CFU/m^3, and after disinfection was 58.9 ± 29.8 CFU/m^3, there was a statistically significant difference ($p < 0.05$). Similarly, before disinfection, the concentration of bacteria in the inpatient room was 424.7 ± 247.2 CFU/m^3, but after disinfection, the average density was 89.1 ± 86.6 CFU/m^3. However, there was no statistical significance ($p > 0.05$). For 5% Hydroperoxide - 0.005% Ag$^+$, the average bacterial density before disinfection in the surgery room was 196.6 ± 130 CFU/m^3 and 110.1 ± 105.1 CFU/m^3 after disinfection. The difference was statistically significant ($p < 0.05$). In the inpatient room, before disinfection by 5% hydroperoxide - 0.005% Ag$^+$, the bacterial density was 398.4 ± 190.3 CFU/m^3 and then reduced to 183.5 ± 154.4 CFU/m^3 after. The difference was statistically significant ($p < 0.05$).

Our research's average bacterial density in the surgery room was 110.1 ± 73.8 CFU/m^3, significantly lower than the study of Zemachu Ashuro [10], which had an average density of 1075 ± 107.1 CFU/m^3. In our investigation, the average bacterial density in the surgery and inpatient rooms was 157.3 ± 36.3 CFU/m^3 and 424.7 ± 247.2 CFU/m^3, respectively.

The average fungus density on Sabouraud Agar medium before disinfecting the air with silver nano solution was 15.7 ± 192 CFU/m^3. The average fungus density was 7.8 ± 14.1 CFU/m^3 lower after disinfection. However, this difference was not statistically significant ($p > 0.05$). The average density of gram-negative bacteria on Macconkey Agar medium before sterilization was 7.8 ± 11.3 CFU/m^3. However, after sterilization, there were no bacteria on Macconkey Agar agar, which was statistically significant ($p < 0.05$). This means nanosilver performs well in disinfecting gram-negative bacteria.

On Blood Agar medium, the average bacterial density before sterilization was 176.1 ± 143.2 CFU/m^3, and after sterilization was 41.5 ± 42.9 CFU/m^3; this difference was statistically significant ($p < 0.05$). For 5% Hydroperoxide - 0.005% Ag, the bacterial density on Sabouraud Agar medium before disinfection was 35.9 ± 36.5 CFU/m^3 and was reduced to 12.3 ± 14.4 CFU/m^3 after disinfection; this difference was statistically significant ($p < 0.05$). On the MacConkey Agar medium, the density was decreased after disinfection, but there was no statistical significance ($p > 0.05$). On Blood Agar medium, the density decreases from 286 ± 272.8 CFU/m^3 to 134.6 ± 102.6 CFU/m^3. The difference has statistical significance ($p < 0.05$). On the overall medium, the bacterial density has also declined hugely from 421.3 ± 397.4 CFU/m^3 to 183.5 ± 154.4 CFU/m^3. The difference has statistical significance ($p < 0.05$).

The data shows that after disinfecting with nanosilver, the average bacterial density in the surgery room was 39.3 ± 20.3 CFU/m^3, which was lower than after disinfecting with 5% Hydroperoxide - 0.005% Ag$^+$, which was 110.1 ± 105.1 CFU/m^3. In the clinic room, nanosilver gave the result of 15.7 ± 0 CFU/m^3 below the result of 5% Hydroperoxide - 0.005% Ag$^+$ of 283.1 ± 66.7 CFU/m^3 of 283.1 ± 66.7 CFU/m^3. The results of bacterial density of minor surgery and inpatient room show the same comparison result, which is that bacterial density after nanosilver aerosol disinfection was lower than bacterial density after 5% Hydroperoxide - 0.005% Ag$^+$ aerosol disinfection. However, the abovementioned difference has no statistical significance ($p > 0.05$).

V. CONCLUSIONS

The study's findings revealed that both nano sliver solution and 5% Hydroperoxide - 0.005% Ag^+ solution reached the standard of V. Omelanski about aerosol disinfection. The percentage of good, qualified aerosol disinfection rooms of nanosilver (69.23%) was greater than 5% Hydroperoxide - 0.005% Ag^+ solution (15.39%). In conclusion, nanosilver can be an alternative method for   5% Hydroperoxide - 0.005% Ag^+ solution in the field of aerosol disinfection.

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