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Antimicrobial effects of harmel smoke on microbial load of hospital wards

Antimicrobial effects of harmel smoke

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Abstract

Aim: Harmel (Peganum harmala L) is one of the most important herbal plants due to its antimicrobial effects. It is used traditionally for treating some diseases. This study aimed at comparing the antimicrobial effect of Harmel smoke and Nocospray solution on the microbial load of hospital wards. Material and Method: From each ward of an educational hospital, two trolleys were collected. First the samples were collected from the trolleys' surfaces and were cultured in Blood Agar and McConkey Agar culture media. Then, they were divided into two equal groups, half of them were disinfected by Harmel smoke and the other half with Nocospay solution. The results were analyzed using SPSS16. Results: The results showed that Nocospray has a stronger antibacterial effect on Staphylococcus and gram-positive bacteria, while Harmel smoke was more effective in gram-negative bacilli. Discussion: Considering that Harmel smoke has the same antimicrobial properties as Nocospray does, it is an effective agent that can be used for disinfecting medical settings.

Keywords

Harmel Smoke; Disinfection; Sterilization; Nosocomial Infection; Medicinal Herb

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Introduction

Nosocomial infection is one of the major issues that can lead to deaths in hospitalized patients. It can also exacerbate illnesses and risks of death in patients due to increased duration of hospitalization. Thus, it increases healthcare costs and has a significant impact on the economy of treatment [1]. This issue is increasingly threatening all hospitalized patients [2]. Since 1970, nosocomial infections have been responsible for more than 800,000 deaths per year. In the United States, hospitals were the eleventh leading cause of death and more than 14 percent of hospital mortalities [3]. The prevalence of nosocomial infections is directly related to the environmental hygiene of hospitals [2]. According to the studies, using contaminated instruments is the most important way of transmitting these infections [4]. According to the studies, various pathogens exist in hospital environment and equipment which are the normal flora of the skin, respiratory system, and digestive tract such as E. coli, Staphylococcus aureus, Pseudomonas, Klebsiella, and fungi. In some investigations, the number of grown colonies were higher than the average standard [5, 6]. Currently, there are disinfection methods and materials which are used in hospitals such as Ultraviolet radiation, fog-making by nebulizer, and also disinfectants such as hydrogen peroxide, glutaraldehyde, formaldehyde, ethylene oxide, Surfosept, sodium hypochlorite, and a newer brand named Nocospray [7, 8]. Although each of these substances can play a role in disinfecting the hospital wards, but environmental and human complications of using these chemical substances are inevitable [9, 10]. Therefore, using herbal ingredients has increased in recent years. The vast majority of drugs are synthetic, but at least one-third of them are produced after extraction of plants [11]. In the recent century, bacterial infections and resistance to antibiotics have been a serious threat to human health. Therefore, developing new antimicrobial compounds with minimal side effects is a critical issue. Protecting themselves against pathogens, plants have to create specific defense mechanisms and synthesize antimicrobial compounds which are always a valuable source for production of antimicrobial compounds [12].

Many of ancient drugs are still used in the same ancient forms, including medicinal smoke. Using medicinal smoke was common in more than 50 countries. It is believed that the smoke has the therapeutic effects of a drug, even stronger and faster than regular drug forms. Harmel smoke is one of these medicinal smokes [13]. Peganum harmala L., locally called Espand, is one of the most important medicinal plant species due to its antimicrobial properties and is traditionally used in the treatment of infectious diseases. Harmel contains antimicrobial substances such as Flavonoids and beta-carboline alkaloids, which are found in root, seeds, and callus of the plant. In traditional medicine of Central Asian countries (Iran, Turkey, Pakistan, Afghanistan, and Yemen), extract of different organs of harmala plant is used for different purposes such as repelling the evil eye, disinfecting the air, increasing milk secretion, excreting parasites, disposing of intestinal worms, treating rheumatism using their anti-inflammatory effect, and sexual enhancement, and also as a strong painkiller [13].

Various studies have proven the antimicrobial effect of Harmel smoke, including reduction of microbial load of Pseudomonas aeruginosa and Staphylococcus aureus in laboratory environment [1]. Reduction of Spore-forming and non-spore-forming Gram-positives, yeast, and filamentous fungi in factory farms, and also the antimicrobial effect on biofilms of Enterococcus faecalis [14, 15]. However, the antimicrobial effect of Harmel smoke in the clinical environment has not been studied. Therefore, in this study investigated the effect of Harmel smoke from Peganum harmala seeds on microbial load of the hospital environment and compared its effect with that of Nocospray, which is a common disinfectant used in burn wards and operating rooms of the hospitals.

Material and Method

The samples in this study were surfaces of the trolleys collected from different wards of an educational hospital in Arak, Iran. In this investigation, a total number of 57 trolleys were studied within two days. In the first day, 30 trolleys were collected and randomly divided into two equal groups. They were numbered and moved to two separated and isolated rooms. Before performing any disinfection, the samples were taken from the surfaces of trolleys in an area of two square centimeters, using sterile swabs moistened in sterile saline. The swabs impregnated onto trolley surfaces were treated directly on Blood Agar and MacConkey Agar culture media. Between the two groups of trolleys, one was randomly selected for disinfecting by Harmel smoke, and the other by Nocospray, which is commonly used as disinfection solution in hospitals. Nocospray disinfection process lasted one hour. A fog-making device sprayed this antiseptic solution for 30 minutes. Another 30 minutes was considered as retention time for disinfecting. In order to disinfect through Harmel smoke, 500 grams (15 grams per each cubic meter of the isolated room) of Harmel seed was purchased from a grocery and was smoked for 30 minutes by the flame. Another 30 minutes was considered to let the smoke entirely coat the trolleys' surfaces. Finally, after sampling from both groups, as performed in the first stage, the culture media were transferred to an incubator with the temperature of 37°C.

On the second day, 27 trolleys from the same wards of the educational hospital were studied. They were transferred to two separate rooms, and after numbering, the samples were collected. This time before disinfecting the trolleys by Harmel smoke and Nocospray, trolley surfaces were soaked and washed with the foam produced from a detergent. Then, they were randomly separated into two groups, 13 trolleys were collected to be disinfected by Harmel smoke and 14 by Nocospray. Disinfecting surfaces and collecting the samples were performed on the first day and the samples were cultured directly on Blood Agar and McConkey Agar culture media and transformed into an incubator with the temperature of 37°C.

After 72 hours of incubation, bacteria colonies appeared on the culture media. The colonies were counted and studied to determine the genus and species of the bacteria. Gram staining technique was performed to identify gram positivity or negativity of the bacteria. Differential tests for Gram-positive bacteria were performed including catalase test, Mannitol salt agar, and antibiogram test. Following tests were performed to identify Gram-negative bacteria: Triple Sugar Iron test, oxidative-fermentative test, and oxidase. Finally, the results were reported.

Results

In this investigation, 57 Trolleys of different hospital wards were studied, and the results are summarized in the tables and figures below.

Table 1. illustrates the growth of bacteria before disinfecting. Except in the case of Gram-negative bacilli, there is no statistically significant difference between the two methods.

Table 2. shows the growth of bacteria before disinfecting. Except in the case of Gram-negative bacilli, there is no statistically significant difference between the two methods.

Table 3. shows the growth of microorganisms after disinfecting by Harmel smoke and Nocospray, and there is no statistically significant difference between the two methods. It means that the growth of all bacteria, except Gram-positive bacilli, has been dramatically reduced.

Table 4. shows that there is no significant difference between the growth of bacteria after disinfection by Harmel smoke and Nocospray. This means that Harmel smoke has the same disinfectant property as Nocospray does.

Table 1. Comparison of positive cultures before disinfecting by Nocospray and Harmel smoke

	Number of Samples	Disinfection Method	Staphylococcus	Gram-positive Bacilli	Micrococcus	Gram-Negative Bacilli	Pseudomonas	Streptococcus
Positive culture	15	Harmel Smoke	2	15	0	9	0	0
	15	Noco-spray	5	15	1	2	0	1
Mann-Whitney test			0.2	1	0.31	0.009	1	0.31

Table 2. Comparison of positive cultures before washing and disinfecting with Harmel smoke and Nocospray

	Number of Samples	Disinfection Method	Staphylococcus	Gram-positive Bacilli	Micrococcus	Gram-Negative Bacilli	Pseudomonas	Streptococcus
Positive culture	13	Washing + Harmel smoke	12	10	6	7	1	2
	14	Washing + Nocospray	10	12	4	1	3	3
Mann-Whitney test			17/0	0.56	35/0	009/0	0.32	0.69

Table 3. Comparison of positive cultures before disinfecting by Nocospray and Harmel smoke

	Number of Samples	Disinfection Method	Staphylococcus	Gram-positive Bacilli	Micrococcus	Gram-Negative Bacilli	Pseudomonas	Streptococcus
Positive culture	15	Harmel Smoke	1	15	0	0	0	0
	15	Nocospray	3	15	0	1	0	0
Mann-Whitney test			0.29	1	1	0.31	1	1

Table 4. Comparison of positive cultures after washing and disinfecting with Harmel smoke and Nocospray

	Number of Samples	Disinfection Method	Staphylococcus	Gram-positive Bacilli	Micrococcus	Gram-Negative Bacilli	Pseudomonas	Streptococcus
Positive culture	13	Washing + Harmel smoke	7	5	2	0	0	0
	14	Washing + Nocospray	3	4	1	0	0	0
Mann-Whitney test			087/0	0.59	0.5	1	1	1

Discussion

In this study, 57 samples from different wards of an educational hospital were investigated in two stages: washing stage and no-washing stage. The results showed that before disinfection, there is no statistically significant difference in the growth rate of Gram-negative bacilli, Staphylococci, Micrococcus, Streptococcus, and Pseudomonas between the two groups of Harlem and Nocospray ($P<0.05$). However, the results show that the growth rate of gram-negative bacilli was significantly higher in Harmel smoke group ($p=0.009$). In no-washing stage, the culture results of bacteria indicate that neither Harlem group nor Nocospray group is effective on gram-positive and gram-negative bacteria. Although Harlem smoke seems to be more effective on gram-negative bacteria, this difference was not statistically significant. At this stage, the growth rate of Pseudomonas, Streptococcus, and Micrococcus was poor, but in washing stage, both methods of disinfection via Harmel smoke and Nocospray are effective on staphylococcal, gram-positive and Micrococcus. In this regard, although Nocospray seems to be more effective than Harmel smoke, but the difference was not statistically significant. Also in this stage, both methods had antimicrobial effects on gram-negative bacilli as no growth was detected on culture media after incubation. There is no similar study investigating Harlem smoke effects in clinical settings. In their study, Parvin et al. (2010) compared the effects of Harmel and Dung smoke and that of straw smoke via disk diffusion test method. Their results showed that Harmel smoke had antimicrobial effects on Staphylococcus aureus, and the diameter of the of inhibition zone increased with an increase in smoking process [1]. Parvin et al. found that Harmel smoke had antimicrobial effect on Gram-positive bacteria with or without spores, yeast, and filamentous fungi, but it was partially effective for gram-negative bacteria at high doses (8 hours of smoking); while the present study showed that Harmel smoke is more effective on Gram-negative bacilli [15]. The researchers suggest that Harmel smoke can be used as a kind of indoor disinfectant agent [15]. Most studies have focused on Harmel seed extract, for instance, the study of Mazandarani et al. (2009) showed that Bacillus licheniformis bacteria has the most sensitivity to Harmel extract, but Staphylococcus aureus, Staphylococcus epidermisis, Micrococcus loteus, Salmonella typhimorium, and Shigelladysentria, each has the sensitivity of 50 percent. Although Gram-positive bacteria were more sensitive than Gram-Negative bacteria, the difference was not statistically significant [13]. The results of the study showed that Harmel smoke can be as effective as the commercial Nocospray solutions, used as

a common disinfectant in the operation rooms and bath burn units via fog-making instruments. The Mann-Whitney test results show there is no difference between the two methods of disinfection, Harmel smoke, and Nocospray, on Staphylococcus, Gram-positive and Gram-negative bacteria ($P<0.05$). Comparing the two methods of disinfection before and after washing with detergent, there was a significant difference in the growth rate of Gram-negative bacilli. Hence, rinsing the equipment with detergents before disinfection increases the effectiveness of disinfecting.

Conclusion

The results of the present study showed that antimicrobial effect of Harmel smoke is the same as that of the commercial Nocospray solution. However, rinsing the equipment increases the disinfection effect in both methods. So, using Harmel smoke is recommended as a disinfectant in the absence of any contact with the patient and its attendant. Using this method certainly costs less and is more environmentally friendly than other kinds of chemical substances. Further research in this area is recommended.

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