Humidifiers for oxygen therapy: what risk for reusable and disposable devices?

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Introduction. Nosocomial pneumonia accounts for the vast majority of healthcare-associated infections (HAI). Although numerous medical devices have been discussed as potential vehicles for microorganisms, very little is known about the role played by oxygen humidifiers as potential sources of nosocomial pathogens. The purpose of this research was to evaluate the safety of the reuse of humidifiers by analysing the rate of microbial contamination in reusable and disposable oxygen humidifiers used during therapy, and then discuss their potential role in the transmission of respiratory pathogens.

Methods. Water samples from reusable and disposable oxygen humidifiers were collected from different wards of the University Hospital of Messina, Italy, where nosocomial pneumonia has a higher incidence rate due to the “critical” clinical conditions of inpatients. In particular, we monitored the Internal Medicine and Pulmonology wards for the medical area; the General Surgery and Thoracic and Cardiovascular Surgery wards for the surgical area and the Intensive Care Unit and Neonatal Intensive Care Unit for the emergency area. The samples were always collected after a period of 5 days from initial use for both types of humidifiers. Samples were processed using standard bacteriological techniques and microbial colonies were identified using manual and automated methods.

Results. High rates of microbial contamination were observed in samples from reusable oxygen humidifiers employed in medical (83%), surgical (77%) and emergency (50%) areas. The most relevant pathogens were Pseudomonas aeruginosa, amongst the Gram-negative bacteria, and Staphylococcus aureus, amongst the Gram-positive bacteria. Other pathogens were detected in lower percentage. The disposable oxygen humidifier samples showed no contamination.

Conclusions. This research presents evidence of the high rate and type of microbial contamination of reusable humidifiers employed for oxygen therapy. These devices may thus be involved in the transmission of potential pathogens. It could be important, for the prevention of nosocomial pneumonia, to replace them with single-use humidifiers for which the absence of microbial contamination has been confirmed.

Respiratory infections are one of the major nosocomial infections. The etiology of nosocomial pneumonia is determined by the length of hospitalization [8]. Early-onset nosocomial pneumonia, which usually occurs within the first 4-5 days of hospitalization, is generally caused by community-acquired pathogens, such as Streptococcus pneumoniae, methicillin-sensitive S. aureus, Haemophilus influenzae and Moraxella catarrhalis. Conversely, late-onset nosocomial pneumonia (usually after 5-6 days of hospitalization) is generally caused by P. aeruginosa, Acinetobacter spp., methicillin-resistant S. aureus, etc. P. aeruginosa and the Enterobacteriaceae are the most common pathogens that cause nosocomial pneumonia after 10 or more days of hospitalization [7]. In order to reduce the risk of infection for patients, it is necessary to eliminate all potential sources of respiratory pathogens. Oxygen humidifiers are commonly utilized in hospitals, because the oxygen used is a dry and irritating gas that, if poorly humidified, causes lesions of the respiratory mucosa [9]. In past years, some studies have been concerned with the role of humidifiers used for oxygen therapy as sources of nosocomial pathogens causing respira-
tory infections [10-12]. Moiraghi et al. reported 5 cases of fatal pneumonia caused by *Legionella pneumophila*, as a result of nebuliser therapy that was administered using contaminated humidifiers [13]. Bacteria may derive from the oral cavity and distal airways of a patient using the device, or from the contamination of the water involved. On this last point, extensive evidence was given that some potentially pathogenic microorganisms, such as *P. aeruginosa*, are able to grow in distilled water [14]. Other potential sources of device contamination are associated with the inappropriate maintenance of medical devices by healthcare workers. In order to avoid this, the Centers for Disease Control and Prevention (CDC) published a set of guidelines for the prevention of nosocomial pneumonia. The CDC highly recommends the diligent use and careful maintenance of reusable wall-mounted oxygen humidifiers [15, 16]. However, meticulous daily maintenance of medical devices is a time-consuming and laborious task to fulfill in the clinical environment. A possible solution to this problem might be to use sterile disposable oxygen humidifiers. Some studies have demonstrated that disposable humidifiers are safer for patients than the commonly-used reusable ones [17, 18]. Indeed, evidence has been presented that disposable humidifiers can be safely used for 30 days [19, 20]. Another study conducted by Meehan et al. has shown that disposable humidifiers could remain sterile for up to 77 days [21]. The clinical and economic impact of nosocomial pneumonia is an especially important topic for Public Health policies and for the prevention of hospital-acquired respiratory infections.

The purpose of this research was to evaluate the safety of the reuse of humidifiers after their disinfection by determining the rate of microbiological contamination of reusable versus disposable oxygen humidifiers used in medical, surgical and emergency care units at ‘G. Martino’ University Hospital, Messina and to show their potential role in the transmission of important nosocomial pathogens that cause hospital-acquired pneumonia.

**Materials and methods**

The study was carried out over a six-month period (January-June 2015) in the most at-risk medical, surgical and emergency care units of the ‘G. Martino’ University Hospital, Messina. We focused our attention on wards where the incidence of HAI was more frequent than in others wards. Specifically, we monitored the Internal Medicine and Pulmonology wards for the medical area; General Surgery and Thoracic and Cardiovascular Surgery wards for the surgical area and Intensive Care Unit and Neonatal Intensive Care Unit for the emergency area. In the first step of the study, water samples from reusable wall-mounted oxygen humidifiers were randomly collected after 5 days of use: 100 samples were obtained from the medical area (50 from Internal Medicine and 50 from Pulmonology), 100 from the surgical area (50 from General Surgery and 50 from Thoracic and Cardiovascular Surgery), and 100 from the emergency area (50 from Intensive Care Unit and 50 from Neonatal Intensive Care Unit). In the second step, water samples from sterile disposable oxygen humidifiers were collected after the same amount of time used for reusable humidifiers. Water samples from 50 disposable oxygen humidifiers were gathered from each ward following the same method that was used in the first step of the research.

In order to obtain the samples, 3 ml of water were collected from the humidifiers using a sterile Pasteur pipette and transferring it to a sterile test tube containing nutrient broth (BBL Nutrient Broth, BD). Immediately after, water samples were transported to the laboratory and incubated at 37°C for 24-48 hours. We considered contaminated each sample that showed bacterial growth. From positive samples, we prepared further subcultures on different growth media: Blood Agar (bioMérieux) was used as a universal medium; Mannitol Salt Agar (Oxoid) was used for the isolation of *Staphylococcus spp*; MacConkey Agar (bioMérieux) was used for the isolation of Gram-negative bacteria; Enterococcosel Agar (bioMérieux) for *Enterococcus spp*; Sabouraud Agar (bioMérieux) for mycetes. Following that, the identification of microorganisms grown in subcultures was carried out using manual biochemical methods (API Identification System, bioMérieux: API STAPH for *Staphylococcus spp*; API 20 NE for non-Enterobacteriaceae Gram-negative bacteria; and API 20 E for Enterobacteriaceae Gram-negative bacteria) and automated biochemical ones (VI-TEK, bioMérieux).

**Results**

In the Internal Medicine ward, 41 out of 50 (82%) samples from reusable wall-mounted oxygen humidifiers showed evidence of microbiological contamination. Similarly, in the Pulmonology ward, 42 out of 50 (84%) samples were contaminated. The total positivity of the medical area was 83% (83 out of 100). In the General Surgery ward, 40 out of 50 (80%) samples provided evidence of microbial contamination and in the Thoracic and Cardiovascular Surgery ward, 37 out of 50 (74%) samples were contaminated. The total positivity of the surgical area was 77% (77 out of 100). Finally, in the Intensive Care Unit and Neonatal Intensive Care Unit, 20 out of 50 (40%) and 30 out of 50 (60%) samples showed evidence of microbiological contamination, respectively. The total positivity of the emergency area was 50% (50 out of 100). The microorganisms detected in the contaminated samples were: *P. aeruginosa*, *Klebsiella pneumoniae*, *Acinetobacter baumannii*, *Serratia marcescens*, *Serratia liquefaciens*, *Proteus mirabilis*, *Citrobacter freundii*, *Stenotrophomonas maltophilia*, *Chryseobacterium indolgenes*, *Vibrio vulnificus* and *Ochrobactrum anthropii* amongst the Gram-negative bacteria; *S. aureus*, coagulase-negative staphylococci, *Enterococcus spp*.
and *Bacillus spp* amongst the Gram-positive bacteria. Tab. 1 shows the percentages of samples that resulted positive for each microorganism. Sometimes, various microorganisms have been found simultaneously on the same sample, so that positivity for a single microorganism is greater than the positivity of single samples. Conversely, samples from disposable oxygen humidifiers, which were collected in the second stage of the research, showed no evidence of any form of contamination.

**Discussion and conclusions**

Initially, the study was conducted using a large number of water samples from reusable wall-mounted oxygen humidifiers obtained from different wards. Special attention was given to those samples collected from wards where oxygen therapy is often delivered to patients with various pathologies (*e.g.* COPD, asthma, cardio-vascular diseases, *etc.*). The highest percentage of humidifiers that yielded positive results for the presence of one or more bacterial pathogens was recorded in medical areas with similar positivity percentage in the two wards assessed (Internal Medicine and Pulmonology). The surgical area showed a slightly lower rate of contamination, while the emergency area was the least contaminated. Looking at the individual departments, the highest percentage of positivity was found in the Pulmonology ward, while the lowest one was found to be in the Anaesthesia and Intensive Care ward. These results have been shown despite the existence of a hospital procedure that provides for proper maintenance of these devices including: their handling by healthcare workers with disposable gloves, disinfection with sodium hypochlorite 5% for 30 minutes, rinsing with sterile water and drying with a sterile cloth. These results are probably due to several factors, particularly a larger number of patients are generally hospitalized in medical and surgical care units than are in emergency care ones. For this reason, the same reusable oxygen humidifiers were used more frequently. Secondly, high rates of microbiological contamination are probably associated with the inappropriate maintenance

| Tab. 1. Percentage and number of microorganisms detected in the various wards. |
|-----------------------------------|------------------|------------------|------------------|------------------|------------------|------------------|
|                                  | **MEDICAL AREA** | **SURGICAL AREA** | **EMERGENCY AREA** |
|                                  | Internal Medicine | Pneumology | General Surgery | Thoracic-Vascular Surgery | Intensive Care Unit | Neonatal Intensive Care Unit |
| **GRAM-negatives**               |                  |                |                 |                            |                  |                                |
| *Pseudomonas aeruginosa*         | 56% (28)         | 50% (25)       | 58% (29)        | 26% (13)                   | 10% (5)           | 20% (10)          |
| *Acinetobacter baumannii*        | 2% (1)           | 2% (1)         | 6% (3)          | 8% (4)                     | 8% (4)            | 6% (3)            |
| *Klebsiella pneumoniae*          | 4% (2)           | 14% (7)        | 0               | 0                          | 4% (2)            | 0                |
| *Serratia marcescens*            | 14% (7)          | 14% (7)        | 4% (2)          | 0                          | 4% (2)            | 0                |
| *Serratia liquefaciens*          | 2% (1)           | 12% (6)        | 0               | 0                          | 0                | 0                |
| *Proteus mirabilis*              | 12% (6)          | 6% (3)         | 26% (13)        | 0                          | 0                | 0                |
| *Citrobacter freundii*           | 6% (3)           | 2% (1)         | 10% (5)         | 4% (2)                     | 0                | 0                |
| *Chryseobacterium indologenes*   | 2% (1)           | 2% (1)         | 0               | 4% (2)                     | 0                | 0                |
| *Vibrio vulnifucus*              | 0                | 0              | 0               | 0                          | 6% (3)            | 0                |
| *Ochrobactrum anthopii*          | 0                | 0              | 0               | 0                          | 6% (3)            | 0                |
| *Stenotrophomonas maltophilia*   | 0                | 2% (1)         | 0               | 0                          | 0                | 0                |
| **GRAM-positives**               |                  |                |                 |                            |                  |                                |
| *Staphylococcus aureus*          | 14% (7)          | 4% (2)         | 12% (6)         | 18% (9)                    | 6% (3)            | 6% (3)            |
| *Coagulase-negative staphylococci* | 18% (9)       | 10% (5)        | 6% (3)          | 8% (4)                     | 8% (4)            | 14% (7)           |
| *Enterococcus spp*               | 18% (9)          | 10% (5)        | 10% (5)         | 28% (14)                   | 6% (3)            | 6% (3)            |
| *Bacillus spp*                   | 6% (3)           | 6% (3)         | 0               | 0                          | 4% (2)            | 0                |
| *Candida albicans*               | 14% (7)          | 2% (1)         | 0               | 4% (2)                     | 6% (3)            | 6% (3)            |
| **Negative samples**             | 16% (8)          | 18% (9)        | 20% (10)        | 26% (13)                   | 60% (30)          | 40% (20)          |

*The sum exceeds the actual total of samples because of the co-presence of multiple germs*
of medical devices for an incorrect application of the sanitization procedure.

In our research, Gram-negative bacteria account for the vast majority of detected microorganisms, with *P. aeruginosa* being the most commonly-isolated microorganism. This may be due to the ability of *P. aeruginosa* to grow and multiply in water. Other Gram-negative rods, such as *A. baumannii* and *K. pneumoniae*, which are known to cause nosocomial pneumonia [22], were isolated with lower frequency. Amongst Gram-positive bacteria, *Staphylococcus spp* were the most commonly-isolated species, followed by the others species.

These results confirm the idea that reusable oxygen humidifiers may be easily contaminated, and that they may play a role in the transmission of potential nosocomial pathogens, especially in those units that provide assistance and care to patients in critical clinical conditions (e.g. immunocompromised individuals, patients with chronic diseases, etc.). Aerosol particles that generate when small water particles mix with oxygen may serve as excellent vehicles in the transmission of microorganisms, for they reach the deep lung immediately after inhalation. Conversely, all sterile disposable oxygen humidifier samples, which were collected in the second stage of the research, confirm that these devices are safe for patients and that it is necessary to substitute reusable oxygen humidifiers with disposable ones. This could be a preventive way to reduce the risk of transmission of potentially pathogenic microorganisms and to minimize the possible development of nosocomial pneumonia.

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**Conflicts of interest:** the authors declare that there are no conflicts of interest.

**Ethical approval:** not required (this study is not a clinical trial and did not involve human subjects).

**Authors’ contributions**

VLF, GBC, RS conceived, designed and coordinated the research. AF, RR and AC, contributed to the acquisition, analysis and interpretation of data. VLF, AF, RR and GBC evaluated the results. VLF, AF and RR wrote the manuscript. All Authors revised the manuscript and gave their contribution to improve the paper. All authors read and approved the final manuscript.

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