

Contamination of nebulisers and surrounding air at the bedside of mechanically ventilated patients

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Background. The delivery of aerosolised medication, as performed by nurses and physiotherapists in intensive care units (ICUs), forms an important component of patient care.

Objectives. To determine the presence of contamination of nebulisers used within a ventilator circuit; to describe the protocol and clinical practice regarding decontamination and storage of these devices; and to identify micro-organisms colonising contaminated nebulisers and the surrounding air at patients' bedsides.

Methods. A cross-sectional multicentre observational study was conducted, including site and equipment sampling to determine contamination. ICU managers were interviewed to determine the decontamination and storage protocols used for nebulisers in their units. Swabs were taken from nebuliser chambers and streaked onto blood agar plates (BAPs). An air sampler was used to collect air samples from the surrounding bedside environment. The BAPs were incubated for bacterial and fungal contamination. Species of colonies observed in these samples were identified.

Results. Sixty-one nebulisers from seven ICUs were sampled (Micro Mist $n=37$; Aeroneb $n=24$). Half of the nebulisers (Micro Mist ($n=19$, 51.4%)); Aeroneb ($n=12$, 50%) and most air samples ($n=60$, 98%) presented with contamination. All participating ICUs reported decontamination and storage protocols, but visual inspection of nebulisers suggested that the protocols were not observed. Nebulisers rinsed with alcohol and left open to the environment to dry had the lowest contamination rates. Coagulase-negative *Staphylococcus* species (spp.) were mostly found in the surrounding air and Aeroneb samples, and *Enterococcus* spp. were mostly found in the Micro Mist nebulisers.

Conclusion. Although decontamination and storage protocols for nebulisers were in place, nebuliser and air contamination was high, possibly due to poor staff adherence.

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Aerosol delivery of pharmacological agents is an important adjunctive therapy frequently used in patient care during mechanical ventilation (MV).^[1] Information on the frequency of use of aerosol drug delivery for patients on MV in South African (SA) intensive care units (ICUs) is currently not available. Nebulisation is the process whereby liquid medications are aerosolised in order to enhance their penetration into the lower respiratory tract of patients with, for example, lower airway obstruction, pulmonary infection, or needing mucolysis of obstructive pulmonary secretions.^[2] A range of aerosol devices is used for the administration of medication to patients during the period of MV. These devices include jet nebulisers (e.g. Micro Mist (Hudson RCI, USA)), vibrating-mesh nebulisers (e.g. Aeroneb (Aerogen, USA)), ultrasonic nebulisers and pressurised metered-dose inhalers used with a spacer.^[1-3] Internationally the Micro Mist nebuliser is mostly used in ICU settings, followed by ultrasonic nebulisers and, more recently, vibrating-mesh nebulisers.^[3-5] Ellis *et al.*^[6] reported that Micro Mist and ultrasonic nebulisers were mostly used in participating private- and public-sector ICUs in Johannesburg during the time of their survey. Conventionally, physiotherapists and ICU nurses are responsible for the administration of nebulised drug therapy to patients on MV.

The study by Ellis *et al.*^[6] is one of the few identified that investigated the prevalence of nebuliser contamination and decontamination and storage protocols for nebulisers used within a ventilator circuit. In this study, nebulisation was mainly performed using Micro Mist

nebulisers, and more than half of these nebulisers (52%) presented with bacterial growth. Contaminated nebulisers stored in sterile drapes on top of the ventilator had higher bacterial concentrations than those that were contaminated but not stored under a drape. None of the public- or private-sector ICUs surveyed in this study had a nebuliser decontamination and/or storage protocol in place.^[6]

Contaminated nebulisers have been linked to the development of hospital-acquired and ventilator-associated pneumonia.^[7,8] Ventilator-associated pneumonia (VAP) is an infection that occurs more than 48 hours after intubation, and represents 86% of pneumonias acquired in the hospital.^[7] Contaminated hospital air and water are also known as environmental reservoirs contributing to the development of nosocomial pneumonia.^[8] The extent of microbial contamination of bedside surfaces and the surrounding air of patients in ICUs in Taiwan was studied. The authors noted that *Pseudomonas aeruginosa* was the most frequently detected and abundant bacterium in the samples collected.^[9] There is currently no information available regarding the association between micro-organisms cultured from contaminated nebulisers used in MV circuits and air samples taken from around the ICU patient's bedside.

The research questions for this study were:

1. What is the presence of nebuliser contamination after use in a ventilator circuit?
2. Are nebuliser decontamination and storage protocols in place and implemented in clinical practice?

3. Is there an association between decontamination practices and nebuliser contamination?
4. Which micro-organisms colonise contaminated nebulisers and the surrounding air at patients' bedsides?

Methods

A cross-sectional multicentre observational study was done. Ethical clearance (ref. no. M120514) was obtained from the Human Research Ethics Committee at the University of the Witwatersrand (Wits). Permission to conduct the study at the respective hospitals was received from the hospital manager or chief executive officer and the specific ICU manager. All hospital managers and unit managers provided written informed consent before data collection commenced. Sixteen hospitals (private and public sector) in Pretoria, SA, were approached for participation in the study. Four private hospitals provided consent. The inclusion criteria for this study were having nebulisers in the ICU at the time of audit that had been used within a ventilator circuit attached to an endotracheal or a tracheostomy tube. Nebulisers of patients nursed in isolation cubicles were excluded from the study owing to known diagnosed infections. The unit audit tool used by Ellis *et al.*^[6] was reviewed and adapted to meet the objectives of this study. The tool assessed information related to the ICU environment, number of staff on duty, presence of nebuliser decontamination and storage protocols and observations made regarding nebuliser storage. Using this tool, unit managers of participating ICUs were interviewed (once-off) to determine the existence of decontamination and storage protocols for nebulisers in their respective units. After the interview, the unit manager indicated which patients were receiving MV at the time, and identified the nebulisers stored at their bedsides. Visual inspection of each nebuliser was done to identify the presence of remaining liquid in the nebuliser reservoir, and to observe the storage procedure.

Nebuliser swabs were collected first. The Micro Mist nebuliser was removed from the oxygen tubing and/or covering and placed on sterile gauze on a sterile workstation. The easy-seal threaded cap was removed and placed on the sterile gauze. The base plate was then removed without touching the sides of the chamber and also placed on the sterile gauze. A sterile swab was dipped into the residual solute within the reservoir of the nebuliser and immediately streaked across a sterile blood agar plate (BAP). This procedure was repeated twice in order to collect two sets of BAPs. If the reservoir was dry or there was less than 2 ml of liquid in the reservoir, 2 ml of 0.9% sodium chloride solution was added to the reservoir. Each nebuliser was reassembled and returned to the patient's bedside in its original position and condition. The Aeroneb nebuliser is manufactured to stay attached within the ventilator circuit,^[10] and only its plug was opened for swabbing.

The Surface Air System (SAS) sampler (SAS International PBI, Italy) was used to collect air samples. The SAS sampler was programmed to sample a constant 200 L of air for each air sample taken.^[11] The aspirating metal head and chamber of the SAS sampler were disinfected with 70% alcohol before each sampling procedure. After the device was air dried, a BAP was inserted. Two air samples were taken at each selected bedside, no more than 1 m away from the ventilator. The BAPs were put in individualised resealable plastic bags and stored upside down in a cooler box for safe transportation to the laboratory for incubation. One air BAP and one nebuliser BAP were incubated at 25°C for 7 days for possible fungal contamination, and the others incubated at 37°C for 24 hours for possible bacterial contamination.

After incubation, the number of colony-forming units was counted on each BAP, and each colony was described according to elevation,

colour, shape, size, surface, margins, density, pigments and the presence of haemolysis. The five most frequently observed colonies cultured from the air, as well as nebuliser, for each hospital were selected for identification. Bacterial isolates were identified using conventional, internationally accepted microbiological techniques, including Gram stain microscopy and biochemical reactions. All Gram-negative organisms were identified on the MicroScan Walkaway 96 (Dade-Behring, USA) using the Microscan Rapid Negative ID Type 3 (RNID3) (Dade-Behring, USA) and API (Biomerieux, France) systems. Gram-positive organisms were identified through various testing algorithms. Colonies were then identified with the API20C AUX system (Biomerieux, France). Gram-positive cocci were tested for catalase production with hydrogen peroxide (Diagnostic Media Products, SA). The Prolex latex agglutination test (Prolab-diagnostics, UK) was used to distinguish *Staphylococcus aureus* from coagulase-negative *Staphylococci*. Catalase-negative organisms were tested on both bile aesculin plates (Diagnostic Media Products, SA) and with the PYR 50 test (Remel, USA) to differentiate between *Streptococci* and *Enterococci*. *Streptococci* and *Enterococci* were not identified further, and thus reported as either *Streptococcus* or *Enterococcus* species (spp).

Statistical analysis

Data from the participating hospitals were pooled and analysed as such to maintain hospital anonymity. Descriptive statistics were used to analyse data, and these are presented as frequencies and percentages. The Fischer's exact test was used to ascertain the association between the contamination of Micro Mist nebulisers and storage protocol, by using the following variables: stored wet, stored in a glove, stored under a sterile drape and stored open to the environment. Statistical significance was set at a *p*-value <0.05.

Results

Ninety-two patients across the participating seven ICUs received aerosol therapy. Sixty-one nebulisers (Micro Mist (*n*=37, 61%); Aeroneb (*n*=24, 39%)) were sampled. The types of ICUs included were cardiac, medical, surgical, trauma, neurology and two mixed units. Fig. 1 outlines the

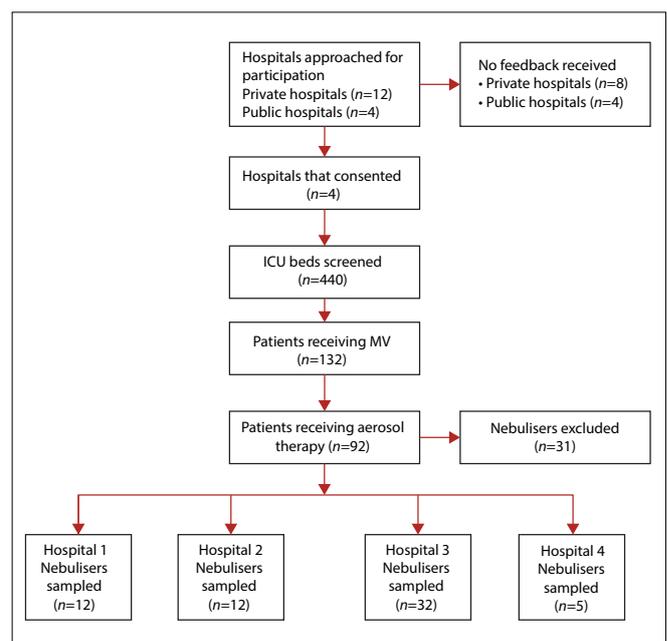


Fig. 1. Flow diagram summarising hospital recruitment procedure and nebulisers and air samples swabbed.

process of hospital recruitment, the number of nebulisers and air samples included and swabbed and reasons for exclusion.

Thirty-one (51%) of the nebulisers swabbed presented with contamination. Both types of nebulisers presented with contamination: Micro Mist ($n=19$, 51%) and Aeroneb ($n=12$, 50%). Contamination was found in the majority of air samples ($n=60$, 98%).

Six decontamination and storage protocols for Micro Mist nebulisers were identified in the seven ICUs included, as in some hospitals, different ICUs used the same protocols. No decontamination and storage protocols were reported for the Aeroneb nebulisers by any of the unit managers. The respective decontamination and storage protocols are outlined in Table 1. Storage protocol 1 stipulated that Micro Mist nebulisers were to be left open to the environment. However, it was observed that Micro Mist nebulisers were not stored on a hook at the bedside, but left open in a petri dish. In protocol 2, used in two ICUs in the same hospital, the Micro Mist nebulisers were not dried before they were stored under a sterile cloth. According to the unit manager, these sterile cloths were replaced every week. In protocol 3, the Micro Mist nebulisers were to be stored inside an acceptor bag; however, no acceptor bag was observed during data collection in that specific ICU.

Using the unit audit tool developed for this study, four types of storage methods (stored in a latex glove, under a sterile cloth, open to the environment or in a paper bag) were defined for visual inspection of each nebuliser in the unit on the day of audit. Although most of the protocols in the ICUs included drying of the Micro Mist nebulisers,

most of them, as well as the Aeroneb nebulisers, were found to be wet during visual inspection. Thirty-three (89%) of the 37 Micro Mist nebulisers, and 20 of the 24 (83%) Aeroneb nebulisers, had retained fluid in their chambers on visual inspection, before the nebulisers were swabbed. Most Micro Mist nebulisers were stored in a latex glove ($n=20$). More than a third of the Micro Mist nebulisers stored in a glove presented with bacterial growth ($n=7$, 35%), and almost half presented with fungal contamination ($n=9$, 47%). Nebulisers stored under a sterile cloth had the highest percentage of bacterial ($n=4$, 44%) as well as fungal contamination ($n=6$, 67%). Nebulisers stored open to the environment resulted in the least bacterial ($n=2$, 29%) and fungal contamination ($n=2$, 29%). One nebuliser was left in the ventilator circuit, and presented with fungal contamination. Five Micro Mist nebulisers were stored connected to the oxygen port of the ventilator. Only one of these presented with both bacterial and fungal contamination. There was no significant association between latex glove storage and bacterial growth ($p=0.72$), or between storage under a sterile cloth and bacterial growth ($p=0.62$). When nebulisers were stored open to the environment, no significant association was observed between the storage method and bacterial growth ($p=0.59$).

For some Micro Mist nebulisers, both bacterial and fungal growth was observed on the same BAP ($n=11$, 58%). On separate BAPs, only fungal ($n=6$, 32%) or bacterial growth ($n=2$, 11%) was observed. For the Aeroneb nebuliser the following contamination was observed: bacterial and fungal growth ($n=9$, 75%), fungal growth only ($n=1$, 8%)

Table 1. Type of decontamination and storage protocols used as reported by the unit managers

Hospital	Protocol	Nebulisers assessed, <i>n</i>		Method of decontamination	Dried nebuliser	Drying method	Method of storage
		Rinsed					
1	1	12	Yes	70% alcohol	Yes	Paper towel	Connected to the oxygen output of the ventilator, stored on a hook at the bedside and open to the environment
2	2	12	Yes	Provac water	No	None	Connected to oxygen, taken apart and left to dry under a sterile cloth
3	3	19	Yes	Wash with Bioscrub once a day	Yes	Paper towel	Stored in acceptor (sterile) bag
3	4	13	No	None	Yes	Paper towel	Stored in a glove connected to the oxygen output
4	5	3	Yes	Saline or sterile water	No	None	Stored in a glove connected to the oxygen output
4	6	2	Yes	Wash with Bioscrub after aerosolisation	Yes	Paper towel	Stored in a glove connected to the oxygen output

Table 2. Microorganisms identified in nebulisers and air samples

Microorganism	Contamination area		
	Micro Mist nebuliser ($n=17$), <i>n</i> (%)	Aeroneb ($n=12$)	Air surrounding bedside ($n=50$)
No growth	7 (41)	3 (25)	15 (30)
<i>Empedobacter brevis</i>	2 (12)	0	0
<i>Stenotrophomonas</i> spp.	1 (6)	0	0
<i>Staphylococcus aureus</i>	0	1 (8)	1 (2)
Coagulase-negative <i>Staphylococcus</i>	0	4 (33)	8 (16)
<i>Enterococcus</i> spp.	2 (12)	2 (17)	2 (4)
<i>Neisseria</i> spp.	1 (6)	0	3 (6)
<i>Pseudomonas stutzeri</i>	0	0	4 (8)
<i>Brevundimonas vesicularis</i>	0	0	1 (2)
<i>Bacillus</i> spp.	0	0	1 (2)
<i>Micrococcus</i> spp.	0	0	1 (2)

and bacterial growth only ($n=2$, 17%). When storage protocol 1 was followed, the least amount of contamination was found ($n=3/12$, 25%). Storage protocols 2 and 3 resulted in the most bacterial and/or fungal contamination: $n=8/12$ (67%) and $n=4/5$ (80%), respectively.

Ten different micro-organisms were identified in the sampled nebulisers and surrounding air (Table 2). Coagulase-negative *Staphylococcus* (CoNS) was the most commonly identified in air samples ($n=8$, 16%), followed by *Pseudomonas stutzeri* ($n=4$, 8%) and *Neisseria* spp. ($n=3$, 6%). Coagulase-negative *Staphylococcus* was the most common organism identified in Aeroneb nebulisers ($n=4$, 33%), whereas *Empedobacter brevis* and *Enterococcus* spp. were the most frequently encountered bacterial contaminants in Micro Mist nebulisers ($n=2$, 12% each). In two instances the same micro-organism was identified in the chamber of a nebuliser and its surrounding air. *Enterococcus* spp. were identified in a Micro Mist nebuliser and its surrounding air, and CoNS was identified in an Aeroneb and its surrounding air.

Discussion

Contamination was found in half of the Micro Mist and Aeroneb nebulisers used within a ventilator circuit, and in most of the samples taken of the surrounding air at patients' bedsides. Visual inspection of the nebulisers stored by patients' bedsides showed that the nebuliser storage protocols of the ICUs were not being consistently observed, which could imply low levels of staff adherence to nebuliser decontamination and storage protocols. This would have to be investigated in more depth in future studies. This study is the first to report the presence of contamination of Aeroneb nebulisers used within a ventilator circuit. The presence of contamination found in Aeroneb nebulisers was similar to that found in Micro Mist nebulisers. This finding was noteworthy, as one would expect less contamination to occur, as the inner portion of the Aeroneb nebuliser does not make contact with the outside environment. This suggests that keeping Aeroneb nebulisers connected in the ventilator circuit does not reduce the risk of contamination. Peckham *et al.*^[12] also expressed their surprise at finding similar rates of contamination between conventional and mesh technology nebulisers used at home by adults with cystic fibrosis.

None of the three public-sector or six private-sector hospital ICUs assessed by Ellis *et al.*^[6] had nebuliser decontamination and storage protocols in place. In contrast, all ICUs that participated in this study had decontamination and storage protocols for Micro Mist nebulisers in place. The decontamination and storage protocols for Micro Mist nebulisers differed between hospitals, and within ICUs in the same hospital. It should be noted that most nebulisers were stored wet, and therefore it seemed that protocols were not being adhered to. Three different rinsing solutions were noted in the protocols identified.

Different cleaning practices of nebulisers used in ICUs and wards are reported in the literature. In a single-centre study performed in India, Jadhav *et al.*^[13] found a reduction in bacterial (87% - 12%) and fungal (75% - 15%) colonisation rates of nebulisers used in ICUs and the wards when nebulisers were washed with soap and distilled water and then disinfected with 70% alcohol. These results were obtained when their staff were educated on performing effective and prompt hand hygiene with alcohol-based hand wash before and after handling the nebulisers.^[13] Another protocol noted the following with regards to cleaning of reusable nebulisers in a hospital setting: clean, disinfect, rinse with sterile water after each use and air-dry.^[14] If a mask or mouthpiece was used during aerosol therapy, these devices would be wiped down with

70% alcohol after each use. The author encouraged good handwashing hygiene practices, and suggested that the inside of the nebuliser should not be touched when left out to dry or during reassembly.^[14]

The importance of good handwashing hygiene and the wearing of gloves when handling a nebuliser should not be ignored as a potential means to lessen nebuliser contamination. In this study, the lowest amount of nebuliser contamination occurred when nebulisers were left open to the environment to air dry, which is congruent with research evidence.^[14] An explanation for this finding is that exposure to light contributes to the inhibition of bacterial growth.^[15] Micro Mist nebulisers that were stored under a sterile cloth presented with a higher percentage of fungal and bacterial contamination, similar to Ellis *et al.*'s^[6] findings. Drying is an important component of decontamination protocols, as devices left wet can result in increased contamination,^[16] as confirmed by this study's findings.

Although identical micro-organisms were identified in both nebulisers and the surrounding air in two instances in the current study, it cannot be assumed that the surrounding air was the only contributor to nebuliser contamination, or vice versa, as results showed that different decontamination and storage methods play an important role in the presence of contamination. Of concern is the relatively high level of contamination found in the air surrounding the ICU beds, as this could potentially pose a number of health risks to ICU staff and to patients' visitors as well. It is known that epidemic pneumonia outbreaks in ICUs occur as a result of contamination of respiratory-therapy equipment, medical aerosols, water and air.^[8] Therefore, staff or visitors with lower immunity might be at greater risk of falling ill. Further research into this aspect of exposure and risk profiling is needed.

Staphylococcus aureus and CoNS, *Enterococcus*, *Stenotrophomonas* and *Neisseria* spp. are known causes of VAP.^[17] *Enterococcus* and CoNS were identified in nebulisers and air samples in this study. A limitation of this study is that no data were collected regarding the patients' infection status at the time of the audit. It is therefore unclear whether the presence of these organisms was as a result of patients having VAP at the time of nebuliser and air assessment, or if the patients were still at risk of developing VAP due to the presence of these organisms. However, when organisms are found in nebulisers and the surrounding air, it can be assumed that the organism would most likely be a contaminant.^[13] The frequency of CoNS cultured in air samples in this study is of concern. Qudiesat *et al.*^[18] reported CoNS as one of the micro-organisms most detected in air samples in government and private healthcare settings, which included ICUs, in Jordan.^[18] In contrast, in Taiwan, *Pseudomonas aeruginosa* was reported as the most frequent and abundant micro-organism found in air samples in ICUs.^[9] Another limitation of the current study is that the ICUs studied were all from the private healthcare setting. Therefore, results cannot be extrapolated to public-sector ICUs.

Recommendations for clinical practice are that nebulisers used within a ventilator circuit should be wiped dry and stored open to the environment, to ensure that the lowest amount of contamination occurs. Unit managers should ensure that ICU nurses are educated on nebuliser decontamination, and that storage protocols are in place. Physiotherapists are responsible for staying abreast of protocols in the ICUs where they work. Furthermore, these protocols need to be implemented, and regular audits of adherence conducted in order to reduce the risk of infection to patients and staff. A longitudinal design to investigate staff adherence and the association between patient diagnosis and micro-organisms identified in nebulisers and the surrounding air at the bedside is recommended.

Conclusion

Nebuliser decontamination and storage protocols were recorded in the participating ICUs, but the presence of air and nebuliser contamination was of concern. The micro-organisms identified in both nebulisers and air samples are associated with the development of VAP. The possible reason for increased contamination appeared to be poor staff adherence to recorded protocols.

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