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Abstract: Ventilator associated pneumonia (VAP) remains a serious problem for critically ill patients. We swabbed nine reusable areas on 20 clinically-used Maquet Servo ventilators from a VA Hospital; shortly after they had been removed from patients and identified bacterial isolates. No bacteria were isolated from most of the samples and of the samples that did grow bacteria, the majority of those had fewer than 10 colonies. The bacteria that were isolated were primarily non-pathogenic Gram-positive skin flora. Of the 20 ventilators swabbed, only one of the cultured bacteria was associated with nosocomial infections: methicillin-resistant *S. aureus*. The most commonly contaminated areas were those most likely to be touched by healthcare professionals: the power button and the screen. The areas in closest proximity to the patients, the inspiratory and expiratory ports were the least often contaminated areas. Overall, very few bacteria were transferred to the reusable areas of the ventilators following clinical use.

Keywords: ventilators, reusable, cleaning, disinfecting, bioburden, ventilator associated pneumonia

Introduction

While mechanical ventilation can be a life-saving and necessary procedure, it often presents complications for patients such as ventilator associated pneumonia (VAP).[1] VAPs result in an increase in hospital stay time and cost for patients.[2,3] Additionally, patients that acquire VAP while in the ICU have a higher mortality rate, although the exact risk of death associated with VAP is still unclear due to confounding variables.[1] While VAP rates have decreased in recent years, at least partly due to changes in clinical practices such as elevating patients at an angle rather than lying horizontally,[4] they still remain a problem.

Many modern ventilators have features that limit cross-contamination from one patient to another, such as frequent changes of disposable tubing and heat and moisture exchangers.[5] However, it is unclear how many bacteria and number of strains are transferred via the surface of the ventilators, often touched by healthcare caregivers directly after touching patients. Due to the danger of nosocomial infections, it is imperative to understand the risk of spreading infection via the ventilator surface. The objective of this study was to swab ventilators after patient use to determine the number and strains of bacteria present on areas of the ventilators that are not single use, and therefore will come into contact with subsequent patients.

Methods

Sampling

Twenty clinically used Maquet Servo ventilators at the VA hospital in Baltimore, MD were swabbed approximately 24-48 hours after being removed from a patient. The machines were covered by an equipment bag and isolated after being removed from the patient until they were swabbed. The patients had been admitted to the hospital in either the surgical intensive care unit or the medical intensive care unit. They were ventilated for an average of 2.49 days due to one of the following issues: to protect the airway following surgery, chronic obstructive pulmonary disease (COPD), cardiopulmonary arrest, respiratory failure, or respiratory insufficiency. Nine components of the ventilator were chosen to swab based on either their proximity of contact with patients or the likelihood of them being touched by healthcare providers (Figure 1.) These components were: 1.

Expiratory inlet, 2. Inspiratory outlet, 3. Exhaust port, 4. Interface buttons, 5. Power button, 6. Knob, 7. Screen,
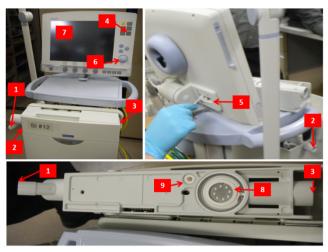

Figure 1: Nine reusable areas of the Maquet Servo Ventilator were swabbed to recover aerobic bacteria. The areas swabbed were: 1. Expiratory inlet, 2. Inspiratory outlet, 3. Exhaust port, 4. Interface buttons, 5. Power button, 6. Knob, 7. Screen, 8. Diaphragm, 9. Exhalation pressure port.

8. Diaphragm, 9. Exhalation pressure port. The expiratory inlet and inspiratory outlet are where the ventilator is connected to disposable tubing, which then directly connects to patients. The exhaust port returns used air to the environment. The interface buttons and knob are present around the screen and allow healthcare providers to interact with the ventilator. The power button turns the unit on and off and is found on the back of the machine. The screen is a touch screen and is often pushed to turn off the alarm. The diaphragm and exhalation pressure port regulate the air pressure within the ventilator.

To collect samples, the ampule within an individually packaged sterile Bactiswab (Remel, Lenexa, KS) was broken to moisten each swab immediately before swabbing one area of the ventilator. The swab was then vigorously swished approximately 10 times in a microcentrifuge tube containing 1 mL of sterile phosphate buffered saline. In order to maximize the amount of sample collected, the swab was then pressed against the internal side of the tube to squeeze out fluid. Samples were plated in triplicate (250 μL/plate) on trypticase soy agar (TSA) plates, incubated aerobically at 37°C, and examined the following day for microbial growth. Each morphologically unique colony type from each plate was restreaked onto a TSA plate and incubated as above.

Identification

Isolates were identified as Gram-positive or -negative using the Gram Staining Set (BD, Franklin Lakes, NJ.) Based on these results, the isolates were then run through the VITEK 2 System (Biomerieux, Durham, NC) on either the Gram-positive or -negative ID card according to manufacturer's instructions. Two samples were identified as *S.aureus*. As methicillin-resistant *S. aureus* (MRSA) is known to cause serious nosocomial infections, we used oxacillin selective

media (Fisher Scientific, Pittsburgh, PA) to determine the cultures' resistance to beta-lactam antibiotics. Five isolates were unidentifiable by the VITEK 2 System and the sixth had confounding results. These samples were subsequently identified using 16S rRNA sequencing technology provided by GE Healthcare (Houston, TX).

Results

Samples were taken from 9 areas on 20 ventilators making 180 samples in total. The majority of the samples (129/180) had no bacteria (Figure 2A); but 19 of the 20 ventilators cultured positive for bacteria on at least one swabbed area (Figure 2B.) Table 1 presents a full list of all the bacteria identified. The areas that most commonly contained bacteria were the power button and the screen with 12 (60%) and 13 (65%) ventilators culturing positive, respectively (Figure 2B.) These two areas are likely the most frequently contaminated because they are often touched by the gloved hand of healthcare providers. The two areas that have the closest proximity to the patients, the expiratory inlet and inspiratory outlet, were amongst the least frequently contaminated having only one (5%) ventilator testing positive for each of the two outlets (Figure 2B.) In general, the

Organism	Number of Instances
Bacillus licheniforms	1
Bacillus megaterium	1
Bacillus pumilusor	1
Bacillus simplex	1
Cronobacter sp.	1
Enterococcus casseliflavus	4
Enterococcus columbae	1
Enterococcus faecium	1
Kocuria rosea	1
Kocuria varians	1
Micrococcus luteus/lylae	4
Pantoea vagans	1
Rhizobium radiobacter	2
Rothia nasimurium	1
Sphingobacterium spiritivorum	1
Sphingomonas paucimobilis	2
Staphylococcus aureus†	2
Staphylococcus capitis	2
Staphylococcus epidermidis	23
Staphylococcus haemolyticus	2
Staphylococcus hominis	11*
Staphylococcus intermedius	2
Staphylococcus lentus	7
Staphylococcus warneri	2*

Table 1: A complete list of the bacteria identified from the 9 areas swabbed from the 20 clinically used ventilators. †

resistant to β-lactam antibiotics (MRSA), * one isolate could not be differentiated between *S. hominis* and *S. warneri* and therefore is represented twice.

ventilators had relatively low contamination levels present shortly after patient use, as the majority of the areas that were
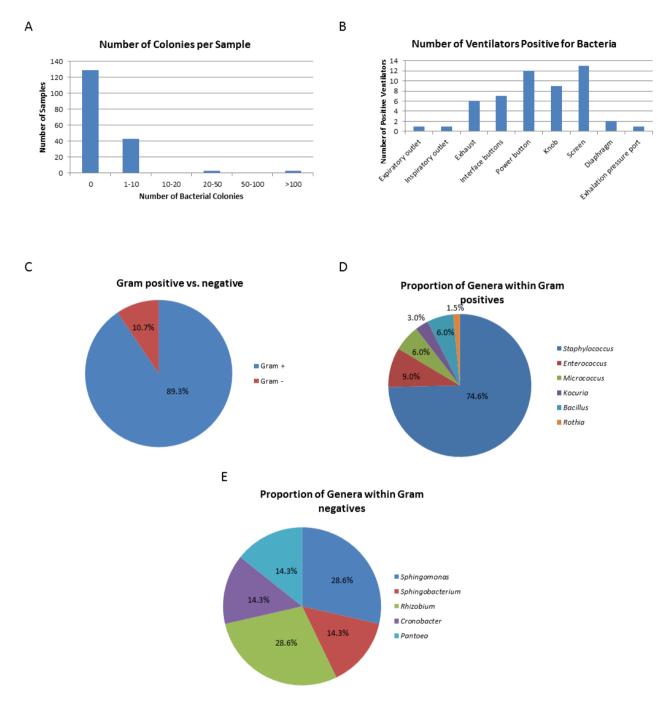
A

Number of Colonies per Sample

B

Number of Ventilators Positive for Bacteria

C

Gram positive vs. negative

D

Proportion of Genera within Gram positives

- Staphylococcus
- Enterococcus
- Micrococcus
- Kocuria
- Bacillus
- Rothia

E

Proportion of Genera within Gram negatives

- Sphingomonas
- Sphingobacterium
- Rhizobium
- Cronobacter
- Pantoea

Figure 2: (A) The number of samples out of a total of 180 that had 0, 1-10, 10-20, 20-50, 50-100, or more than 100 colonies isolated. (B) The number of ventilators that had bacteria isolated from 9 different areas swabbed: Expiratory inlet, inspiratory outlet, exhaust port, interface buttons, power button, knob, screen, diaphragm, and exhalation pressure port. (C) The proportion of bacteria isolated that were Gram-positive and -negative. (D) The proportion of the Gram-positive bacteria isolated in each of the genera represented. (E) The proportion of the Gram-negative bacteria isolated in each of the genera represented.

contaminated with bacteria (43/55) contained fewer than 10 colonies (Figure 2A). Of the bacteria that were present, the majority (89.3%) was Gram-positive (Figure 2C) and 74.6% of these belonged to the *Staphylococcus* genus (Figure 2D.) The rest of the Gram-positive isolates were divided amongst five genera: *Micrococcus*, *Enterococcus*, *Kocuria*, *Bacillus*, and *Rothia* (Figure 2D.) While the Gram-positive bacteria were clearly skewed towards one genus, the Gram-negative bacteria were more evenly split amongst five genera: *Sphingomonas*, *Sphingobacterium*, *Rhizobium*, *Cronobacter*, and *Pantoea* (Figure 2E.) All of the bacteria isolated are characterized as either biosafety level (BSL)-1 or -2 and the majority of the bacteria are associated with common skin flora that do not typically cause infections unless the patient is immunocompromised. One important exception to this observation is that methicillin-resistant *S. aureus* (MRSA) was isolated from the interface and power buttons of one of the ventilators.

Discussion

The ventilators included in this study were used to treat chronic obstructive pulmonary disease (COPD), cardiopulmonary arrest, respiratory failure, respiratory insufficiency, and surgery in order to protect the airway of the patient. Cardiopulmonary arrest and respiratory failure are considered acute respiratory failures, which along with COPD make up approximately 79% of indications for ventilation, indicating that our sample was representative of some of the most common reasons for ventilation.[6,7]

Overall, the ventilators were relatively free of bacterial contamination. The Gram-positive bacteria isolated are primarily commensal skin flora and were principally found on the power button and screen, indicating that the primary mode of contamination is likely the healthcare provider touching the ventilator's interface after touching the patient. After combining the microbiological data with knowledge of normal ventilation protocols, the reason behind the pattern of contamination becomes clear.

When the healthcare professional begins ventilation on a patient they start by using a gloved hand to turn on the power button and then adjust the settings using the interface buttons, the knob, and the screen. They then connect the inspiratory and expiratory outlets to single use tubing, which is then connected to the patient's face. The hospital reports that the tubing is replaced between patients and also when visibly soiled. Once the initial setup is complete, healthcare professionals will enter the room of a patient and interact with the ventilator approximately 50 times a day; the exact frequency at which they enter the room will depend on the patient's individual needs. During each of these interactions, the healthcare professional will put on new gloves when her/she enters the room. They will then touch the patient in order to perform a variety of tasks such as suctioning, listening to breathing sounds, performing chest therapy, and checking vitals. None of these activities require taking the patient off the ventilator as it is a closed system, however they can all cause the machine to register a change in oxygen flow and will therefore cause it to alarm. The healthcare

professional will then touch the screen in order to silence the alarm before continuing to work on the patient. This practice of frequently touching the screen directly after patient contact explains why the screen is one of the most contaminated areas of the ventilator and why the contamination is mostly skin flora. The power button is the second most frequently contaminated and only seems to be touched when a patient is connected or disconnected from the machine. This may be because it is behind a retractable panel, making it more of a challenge to clean. While the retractable panel protects the ventilator from accidentally being turned off after being bumped, it makes the power button more difficult to clean and disinfect.

The outlets connecting tubing are touched when a patient is initially connected to the machine or disconnected as well as once a day in order to change the filters. The hospital reports that when the healthcare professional enters the room to change the filter, he/she will do so with newly gloved hands before touching the patient, indicating that the majority of the interactions with these areas are not preceded by direct contact with the patient. This would be consistent with the fact that most of the outlets connecting tubing to and from patients were found to have no bacteria present. However there was one outlet that connected the machine to the patient which tested positive for *Enterococcus casseliflavus* while another outlet that connected to the machine from the patient tested positive for *Sphingobacterium spiritivorum*. These two instances of contamination are important to note because, while infrequent, they are associated with ventilator components that have a high proximity to the patient. Both *E. casseliflavus* and *S. spiritovorum* are BSL-2 organisms that can cause opportunistic infections.[8,9] Bacteria that are commonly associated with soil, such as *Rhizobium radiobacter and Bacillus pumilus,* were also isolated, indicating that outside soil is also likely a contributing source of contamination. It is important to note that *Bacillus sp*. are difficult to kill because they form spores. While none of these soil bacteria cause infections in healthy individuals, they can do so in immunocompromised patients.[10,11] None of the patients included in this study were immunocompromised, however immunocompromised patients are at an increased risk to develop both infectious and non-infectious pulmonary issues.[12,13] Once immunocompromised patients are ventilated, they have an increased rate of VAP and mortality.[14,15]

The most concerning bacteria isolated from the ventilators were *S. aureus* resistant to beta-lactam antibiotics, or MRSA. MRSA has become an increasingly problematic organism for hospitals to control due to its resistance to multiple antibiotics.[16] While we swabbed 3 ventilators used on patients positively diagnosed with a MRSA infection, only one ventilator tested positive for the bacteria. As we swabbed directly after use on a patient and before the ventilator was subjected to disinfection or sterilization, we can see that the transmission of the bacteria from the patient to the ventilator is low. Additionally, the cleaning/disinfecting/sterilization processes in place are likely working to prevent crossover MRSA contamination given that we did not see any MRSA present on ventilators that did not come directly from isolated

patients. Furthermore, another ventilator isolated due to an infection with a bacteria found in human fluids and cosmetics,[17,18] *Enterobacter gergoviae*, did not test positive for the bacteria in question.

After the ventilator is removed from a patient, the single use components, such as the tubing, are replaced and the reusable areas are cleaned and disinfected. The respiratory therapists wipe down the surfaces of the ventilator (inspiratory and expiratory ports, pressure port, screen, heater, and stand) with Caviwipes; a commercially available cleaning and disinfecting wipe. The expiratory filter is replaced every 24 hours or sooner if soiled. The hospital has also instituted respiratory isolation for patients that have been diagnosed with infections such as MRSA or *Clostridium difficile*. *C. difficile* has the potential to cause devastating gut infections in patients that have recently been treated with antibiotics.[19] Healthcare personnel and visitors must wear clean gloves and scrubs when entering an isolated patient's room and dispose of them when exiting. If a ventilator was used on a respiratory isolated patient, the expiratory cassette is removed and sent to sterile processing for steam sterilization. Bleach wipes are additionally used to clean all external ventilator surfaces, if the ventilator was used on a patient in isolation because of a *C. difficile* infection. It should be noted that one of the limitations of our study is that we are not able to comment on the absence of *C. difficile* in our samples, because we only grew our samples aerobically keeping in mind that all of the surfaces that we sampled are exposed to air and therefore are in aerobic conditions. Additionally, it is possible that there are more bacteria present on the ventilators immediately after use that may die off before we are able to swab them. However, these bacteria would be unlikely to be transmitted to the subsequent patient as they would be dead before the next patient was in contact with the machine. Overall, we have found that there is a low transmission rate of bacteria onto the Maquet Servo ventilators at the Baltimore VA hospital.

Acknowledgments

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