Comparison of Survival of Healthcare Associated Bacteria on Materials used for Making White Coat

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Abstract
Healthcare-Associated Infections (HAIs) are of global concern in this present era and white coats play an important role in the transmission of HAIs. The most common healthcare-associated bacteria are Enterococcus species, Staphylococcus aureus, Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa and Enterobacter species (ESKAPE bugs). These healthcare-associated bacteria are capable of surviving on white coats which could act as fomites in the transmission of HAIs. The purpose of this study was to compare the duration of survival of healthcare-associated bacteria on different materials used for making white coats. Materials like pure cotton, artificial cotton, cotton silk, spun and crepe were cut into swatches of 1cm² size and sterilized by autoclaving. Five different bacteria isolated from clinical specimens were grown on 5 % sheep blood agar and bacterial suspensions were made in sterile physiological saline. The swatches were immersed in bacterial suspension and kept in petri plates at 25°C. The viable counts of bacteria were determined at definite time intervals by surface plate method. The present study shows that among the healthcare-associated bacteria, S. aureus survived the maximum up to 52 days. The duration of survival of S. aureus was significantly longer than P. aeruginosa and A. baumannii (p <0.05). A. baumannii survived only up to 20 days maximum. All the healthcare-associated bacteria significantly survived for the shortest duration of time on crepe. Therefore, crepe could be a better material used for making white coats.

Keywords: Healthcare-Associated bacteria, Survival, White coat materials, Fomites, Gram positive cocci, Gram negative bacilli

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INTRODUCTION

Healthcare-Associated Infections (HAIs) are mainly caused by healthcare-associated bacteria belonging to the “ESKAPE bugs” which include Enterococcus species, Staphylococcus aureus, Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa and Enterobacter species. Different modes of transmission of these bacterial pathogens in the health care settings include direct and indirect contact, inhalation, ingestion and inoculation. Fomites like white coats, gloves, masks and instruments play a significant role in the transmission of healthcare-associated bacteria in the healthcare settings. The white coat is a sign of purity, but still could function as a fomite in the transmission of microorganisms in the healthcare settings.

The duration of survival of different bacteria on white coat may vary based on the type of bacteria, number of bacteria, material used for making white coat, environmental temperature and humidity. Gram-positive bacteria like S. aureus and Enterococcus spp. has known to survive on white coats. Methicillin resistant S. aureus (MRSA) which has the capability of spreading HAIs has also been isolated from the white coats. Among the gram negative bacteria E.coli, P. aeruginosa and A. baumannii are the common healthcare-associated bacteria known to cause HAIs.

Traditionally, the white coats worn by Healthcare workers (HCW’s) are made from cotton, but nowadays many other materials like spun, artificial cotton, cotton-silk and crepe are also used. These healthcare-associated bacteria have survived on materials like cotton, cotton silk, and spun for an extended period of time and can cause HAIs by transferring from gowns to others. With the increased rate of infection caused by these bacteria, studying the inanimate surfaces including the HCW’s attire is also necessary for reducing the rate of HAIs. There is a paucity of studies done on the comparison of survival of healthcare-associated bacteria on white coat materials. Therefore, the purpose of the study was to compare the duration of survival of selected healthcare-associated bacteria on different materials used for making white coat.

MATERIALS AND METHODS

Study setting and study design

The descriptive study was conducted in Department of Microbiology, Kasturba Medical College, Mangalore, during January 2021 to May 2021.

Preparation of white coat materials

White coat materials such as pure cotton, artificial cotton, spun, cotton-silk and crepe were used. Swatches of size 1cm² size were cut from these materials. Then the swatches were sterilised by autoclaving for 15 minutes at 121°C. After sterilisation, the swatches were spreaded on sterile petri plates and dried in incubator at 37°C. After drying, the edges of the plates were covered using a cellophane tape to avoid contamination.

Preparation of bacterial inoculum

The test bacteria such as S. aureus, E. faecalis, E. coli, P. aeruginosa and A. baumannii used in were procured from clinical microbiology specimens and were grown in 5% sheep blood agar, at 37°C for 24hours. These isolates were identified by standard bacteriological methods. The fresh bacterial colonies obtained from the culture were picked using a sterile wire and suspended in sterile physiological saline and turbidity was matched with McFarland 0.5 standard (bacterial count 1.5 x 10⁸ CFU/ml). The bacterial suspension was then

<table>
<thead>
<tr>
<th>Bacteria</th>
<th>Duration of survival of HA bacteria on different white coat materials (Days - Mean ± standard deviation)</th>
</tr>
</thead>
<tbody>
<tr>
<td>S. aureus</td>
<td>Pure cotton: 50.0 ± 4.3</td>
</tr>
<tr>
<td>E. faecalis</td>
<td>Pure cotton: 50.0 ± 3.3</td>
</tr>
<tr>
<td>E. coli</td>
<td>Pure cotton: 43.0 ± 6.2</td>
</tr>
<tr>
<td>P. aeruginosa</td>
<td>Pure cotton: 38.0 ± 2.4</td>
</tr>
<tr>
<td>A. baumannii</td>
<td>Pure cotton: 20.0 ± 0</td>
</tr>
</tbody>
</table>
further diluted 1 in 1000 in sterile physiological saline to get bacterial count $1.5 \times 10^5$ CFU/ml.$^4$  

**Test for survival of bacteria on white coat material**

This method was based on a previously published article but few modifications were made.$^4$ Here, the swatches were soaked in different bacterial suspensions and then they were spreaded on sterile petri dishes. The edges of the plates were covered with cellophane tape to prevent contamination and were kept in room temperature (24-26°C). The swatches were then transferred to 1ml nutrient broth and vortexed once in week. Using a calibrated loop of 4mm internal diameter (0.01ml) the inocula were streaked on 5% sheep blood agar and then colony count was determined. As the colony count reduced, sub-culturing was done on the alternate days. The nutrient broth containing the swatch was incubated at 37°C for 24 hours and then was checked for turbidity. Nutrient broth showing

![Fig. 1. Log viable count of different types of bacteria on pure cotton.](image1)

![Fig. 2. Log viable count of different types of bacteria on artificial cotton.](image2)
no turbidity was subcultured to 5% sheep blood agar to check for bacterial growth. No growth was confirmed as the plate showed no bacterial colony. The mean and standard deviation of the number of days the bacteria survived was calculated and a graph was plotted with number of days on x-axis and log viable count on y-axis.

RESULTS
The present study showed that there was a difference in the duration of survival of different bacteria on white coat. The data from the study showed that *S. aureus* survived the maximum duration for up to 52 days. The duration of survival of *S. aureus* was significantly more when compared with *P. aeruginosa* and *A. baumannii* (p<0.05). Among the gram negative bacteria, *E. coli* survived the maximum up to 47 days. The duration of survival of *E. coli* was significantly more when compared with *P. aeruginosa* and *A. baumannii* (p<0.05). *A. baumannii* survived the shortest duration for up to 15 days on crepe and 20 days on all other materials (Table 1). All HA bacteria have survived the shortest duration of

Fig. 3. Log viable count of different types of bacteria on cotton silk.

Fig. 4. Log viable count of different types of bacteria on spun.
time on crepe. \textit{P. aeruginosa} survived for 17 days and all other bacteria for 15 days on crepe. Fig. 1 - 5 depicts the log viable count of different types of bacteria on different white coat materials like pure cotton, artificial cotton, cotton silk, spun and crepe respectively. The graph shows the reduction in the log viable count of the bacteria.

DISCUSSION

The present study was conducted to compare the duration of survival of healthcare-associated HA bacteria on different materials used for preparing white coats. Materials like pure cotton, artificial cotton, cotton silk, spun and crepe were used for the study. The data from the study shows that the healthcare-associated bacteria could survive up to 52 days. The data was comparable to a previous study which also showed that the duration of survival of these bacteria depend upon the materials used for making white coats.\textsuperscript{10} Among the healthcare-associated bacteria \textit{S. aureus} survived the maximum up to 52 days. The duration of survival of \textit{S. aureus} was significantly more when compared with \textit{P. aeruginosa} and \textit{A. baumannii} (p<0.05). A previous study showed that \textit{S. aureus} has high binding capacity to polyester and very low binding capacity on cotton.\textsuperscript{17} But in this study \textit{S. aureus} survived the longest on natural fibres like cotton and spun (cotton-polyester blend) is seen to be the third material after pure cotton and artificial cotton where it survived for 43 days. Followed by \textit{S. aureus}, \textit{E. faecalis} survived the maximum up to 50 days. This was not consistent with the previous study which suggested that enterococci survived longer than staphylococci.\textsuperscript{3}

Among the gram negative bacteria, \textit{E. coli} survived the maximum up to 47 days. It might be because of the lack of sunlight and low temperature which helped in the growth of gram negative bacteria on white coat materials.\textsuperscript{18} A previous study has also showed that coliform bacteria has good adherence on cotton.\textsuperscript{17} The duration of survival of \textit{E. coli} was significantly longer
than *P. aeruginosa* and *A. baumannii* (*p*<0.05). In the case of *P. aeruginosa*, it survived the maximum up to 38 days in pure cotton, artificial cotton and spun. The least surviving organism was found to be *A. baumannii* and survived the maximum up to 20 days, except on crepe where it survived for up to 15 days and this could be because of the room temperature as the optimum temperature for their growth is 37°C.

Of all these materials, all the healthcare-associated bacteria survived the shortest duration of time on the material crepe. *P. aeruginosa* survived up to 17 days and all other bacteria up to 15 days, suggesting that crepe could be a better material used for making white coat because it sustains the survival of these bacteria for shortest period of time. This could be because of the manufacturing of the material which involves tighter twisting of the yarns or because of the chemical treatment with caustic soda. So overall, the data shows that gram positive bacteria survived longer than gram negative bacteria and it was similar to a previous study.19 This could be because of the rigid body of gram positive bacteria. Fig. 1 - 5 shows the log viable count of different types of bacteria on different white coat materials. The graph shows the reduction of the log viable count. A previous study showed that, even though higher inoculum is used for checking the survival rate of bacteria, the number of bacteria dying in the high inoculum increases to sustain the growth of viable cells so that the viable cells can survive from the nutrients of the dying cells.4 This could explain the decrease in log viable count.

CONCLUSION

The present study showed that the survival of healthcare-associated bacteria on white coat materials depends on the type of bacteria and type of white coat material. *S. aureus* survived significantly more than other bacteria. All the bacteria survived shortest duration of time on crepe. Therefore, crepe could be the material used for making white coat because it sustains survival of these bacteria for the shortest period. Selection of white coat material and frequent laundering of white coats are crucial in controlling HAIs transmitted through white coats.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

AUTHORS’ CONTRIBUTION

Both the authors listed have made a substantial, direct and intellectual contribution to the work, and approved it for publication.

FUNDING

None.

DATA AVAILABILITY

All datasets analysed during the study are included in the manuscript.

ETICS STATEMENT

This study was approved by Institutional Ethics Committee (IEC), Kasturba Medical College, Mangalore, India with Protocol number-IKECKMCMLR-12/2020/401.

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