Original Article

Bacterial flora of the lower respiratory tract during and after a week of tracheostomy

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Abstract

Aims and Objectives: To compare the bacterial flora of the lower respiratory tract during and after a week of tracheostomy. **Study design:** A clinical descriptive study of 21 months duration from Oct 2012 to June 2014. **Study setting:** Department of Otorhinolaryngology and Head and Neck Surgery. Kasturba Medical College Hospital, Mangalore and Government Wenlock Hospital, Mangalore, Manipal University. **Materials and Methods:** All patients on whom an open surgical tracheostomy was done was included. Patients with evidence of lower respiratory tract infection prior to tracheostomy were excluded. A total of two samples of tracheal aspirate were collectedusing a sterile suction catheter from each patient for the study; one during tracheostomy and the other seven days post tracheostomy. These sterile suction catheter tip were send for bacterial culture. **Results:** 130 patients were studied with ages ranging from 28-81 years with a mean age of 57.2 years. Commonest diagnosis in these patients was head and neck malignancy (73.1%) and the commonest indication for tracheostomy being upper airway obstruction (56.2%). The lower airways were found to be colonized in 5.4% of patients according to the culture done during tracheostomy; organisms isolated being *Acenitobacterspps* (3.4%) and *KlebsiellaPneumoniae*(1.5%), all of which were in patient who were previously intubated. Patients developed significantly more colonization post-tracheostomy (113/130) compared to culture done during tracheostomy (7/130). Post tracheostomy cultures taken on seventh post op day showed predominant growth of Gram Negative Bacilli particularly *Pseudomonas aeruginosa*(47.4%), *Klebsiellapneumoniae* (18.5%) and *Acenitobacterspps* (7.7%). **Conclusions:** Post-tracheostomy, lower respiratory tract, which is sterile in normal individuals gets colonized easily with bacteria post tracheostomy commonly by gram negative bacteriae like *Pseudomonas aeruginosa* and *Klebsiellapneumoniae*, Serial tracheal aspirate cultures have to be done to understand the nature of bacteria and treat the infections accordingly.

Key Words: Tracheostomy; Acenitobacterspps; Lower respiratory tract infections; gram negative bacilli; Pseudomonas aeruginosa.

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INTRODUCTION

Tracheostomy is used to describe the creation of a stoma at the skin surface which leads into the trachea[1]. Tracheostomy is one of the oldest surgical procedures known. Today, Tracheostomy plays a pivotal role in airway management. Being a surgical procedure, it is not without complications. In the current era post-tracheostomy lower respiratory tract infection is an important complication[2]. The normal trachea is protected from bacterial colonization, so that the trachea of healthy individuals harbors either no bacteria or oral flora in sparse numbers[3]. These defense mechanisms are partially bypassed following a tracheostomy and direct exposure of the lower airways to the pathogens may occur[4]. The route

of this infection is thought to be endogenous or exogenous[5]. Tracheo-bronchitis is common in patients with tracheostomy and observed rates of 60% have been reported in two adult studies[6,7]. The purpose of this study is to understand the nature of bacterial flora of the lower airways in patients with short term tracheostomy.

MATERIALS AND METHODS

Written informed consents were obtained from all patients or their first degree relatives before the study. Our study, approved by institutional ethics committee is a descriptive time based study of 18 months duration from November 2012 – June 2014 conducted in Kasturba Medical College Hospital, Mangalore, and Government Wenlock Hospital, Mangalore, India.

Patients

All patients in whom an open surgical tracheostomy were performed during the time period were included in the study. Patients with known lower respiratory tract infection prior to the procedure was excluded from the study. This exclusion was based on detailed history including unexplained fever, clinical examination, leucocytosis, neutrophilia and/or chest X-ray findings like consolidation. Leukocytosis is said to be present when the total leucocyte count is more than 9.06x103/cc[8]. In addition, patients on whom tracheostomy was done at night hours were excluded owing to difficulties in transport of samples and storage. Patients who were changed to metal tracheostomy tube during the first tube change were excluded from the study because the sterility of the metal tube could not be confirmed. Thus after applying the exclusion criteria 130 patients (105 males and 25 female subjects) were finally included in the study. The patients were selected consecutively and were included once the inclusion criteria was met. Data including primary diagnosis, indication for tracheostomy and intubation status prior to tracheostomy was noted.

Methods

Open surgical tracheotomy was performed in the major OT in both our institutions under strict aseptic precautions. During the surgery, just after tracheotomy, a sterile suction catheter was introduced into the trachea and tracheal suctioning was done to clear the secretions. Using aseptic precautions, the tip of the suction catheter was cut and placed in a sterile container. The container which was sealed and transported to the microbiology lab for bacteriological analysis. This was labelled as Day 0 culture. Standard post tracheostomy care including regular tracheobronchial suctioning using separate sterile suction catheters, 2 hourly deflation of the cuff for 10 min to reduce the risk of tracheal necrosis and granuloma, Stomal dressing, provision of humidified air were done for all patients post-surgery. Tracheostomy tube change was done every 2-4 days after tracheostomy under strict aseptic conditions. On the seventh post-operative day, tracheal suctioning was done with a sterile suction catheter, its tip cut, put in a sterile container and send for bacteriological analysis. This was the Day 7 culture. Bacterial Cultures were done on McConkeys agar, Chocolate agar and Sheep Blood agar and the isolates were identified using grams staining and standard biochemical reactions.

Statistical Methods

Descriptive statistical analysis has been carried out in the present study. Results on continuous measurements are presented on Mean ± SD and results on categorical measurements are presented in Number (%). Chi-square/ Fisher Exact test has been used to find the significance of study parameters on categorical scale between two or more groups. P value <0.05 were considered statistically significant.

RESULTS

Total of 130 patients were included in the study who underwent tracheostomy during the study period. Mean age of the patients studied was 57.2 years with age range from 28-61 years (Figure 1). This included 105 males and 25 females (Figure 2). Primary diagnoses of the patients undergoing tracheostomy are illustrated in Figure 3. Patients who were diagnosed with head and neck malignancy were by far the main candidates for tracheostomy in our study (73.1%). followed by patients who presented to us with deep neck abscess (5.4%) and bilateral vocal cord palsy (2.3%). Study also included 20 (15.4%) patients from the Intensive care units who were previously intubated for various indications commonest being for cerebrovascular accident. Commonest indication for tracheostomy in our patients was found to be upper airway obstruction (56.2%), almost all of which was found in patients with head and neck malignancy. Others were tracheostomy done for major head and neck surgeries (28.4%) and for patients with prolonged intubation in the Intensive Care Unit (ICU) (15.4%) (Figure 4)

Tracheal aspirate culture

There was no growth in the Day 0 culture from Tracheal suction catheter tip in 94.6 % (n=123) of the patients (Table 1). 5.4% (n=7) of the patients had positive growth, of this majority of the patients had *Acinetobacter spps* (3.9%, n=5) followed by *Klebsiella spps* in 1.5% (n=2) patients. All these 7 patients with positive growth were previously intubated.

In contrast, 113 out of 130 (86.9%) day 7 tracheal suction catheter tip cultures yielded a positive result. With respect to the identity of the bacteriae studied in these positive cultures, majority were gram negative bacilli (n=100)

with 62 (47.7%) samples growing *Pseudomonas aeruginosa* followed by *Klebsiella pneumonia* in 24 samples (18.5%) and staphylococcus aureus in 13 samples (11.5%). Other organisms noted were *Acenitobacter spps* (7.7%, n=10), *Enterobacter spps* (1.8%, n=2), *Citrobacter spps* and *Diphtheroids spps* (Table 2). 17 of the samples failed to grow any bacteria in even after a week of tracheostomy. High statistical significance was noted when comparing the lower respiratory bacteriology between day 0 and day 7 of tracheostomy ($p<0.001$) (Table 3). Where, day 0 culture showed only 7 positive growth, this increased exponentially to 113 positive growths in day 7 culture. Interestingly, Patients with positive day 0 culture showed persistence of the same bacteriae in their day 7 culture.

Table 1: Bacterial growth pattern – Day 0 Culture

Bacterial growth	Frequency (n=130)	Percent
Absent	123	94.6
Present		
Acinetobacter baumanni	7	5.4
Klebsiella pneumoniae	5	3.9
	2	1.5
Total	**130**	**100**

Table 2: Bacterial growth pattern – Day 7 culture

Bacteria	No of patients	percentage
Pseudomonas aeruginosa	62	47.7
Klebsiella	24	18.5
Staphylococcus aureus	13	10
Acinetobacter Baumanni	10	7.7
Diphtheroids	1	0.8
Enterobacter	2	1.5
Citrobacter	1	0.8
No growth	17	13.0
Total	**130**	**100**

Table 3: Comparison of day 0 with day 7 culture

Bacterial growth in tracheal suction catheter tip sample culture	Absent		Present		Wilcoxon signed rank test p value
	Frequency	Percentage	Frequency	Percentage	
Day 0	123	94.6%	7	5.4%	P=0.000<0.001, HS
Day 7	17	13.1%	113	86.9%	

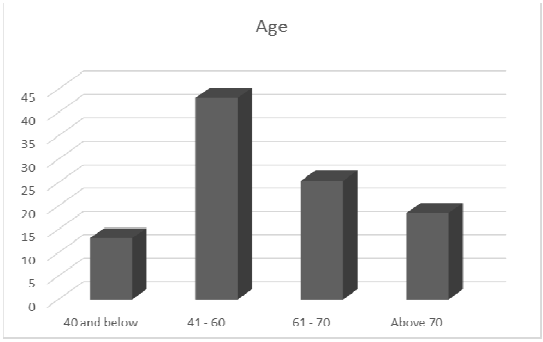

Figure 1

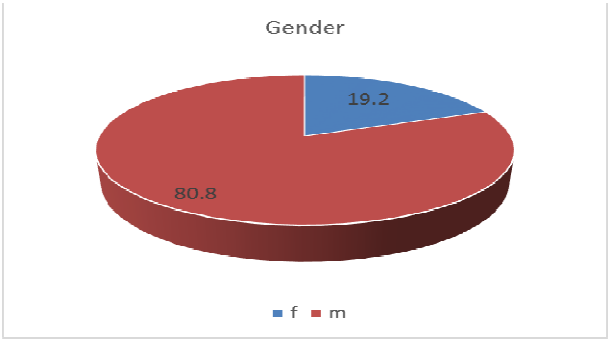

Figure 2

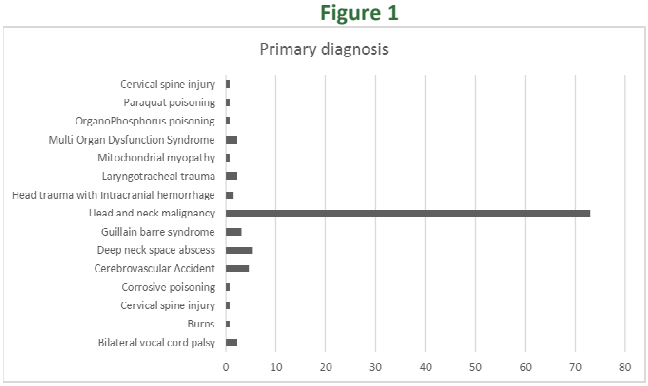

Figure 3

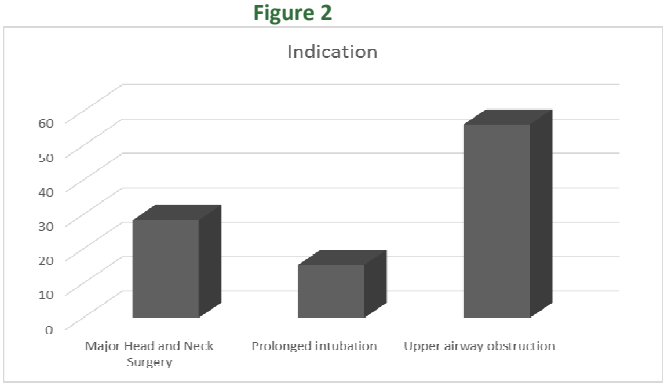

Figure 4

Legend

Figure 1: Age distribution of the patients studied; **Figure 2:** Gender of the patients studied; **Figure 3:** Primary diagnosis of the patients studied; **Figure 4:** Indications for tracheostomy

DISCUSSION

The normal trachea is protected from bacterial colonization, so that the trachea of healthy individuals is considered sterile with no bacterial colonisation[3,9]. Tracheostomy eliminates the filtering mechanisms of the upper airways, reduces the effectiveness of the cough reflex, and interferes with glottic closure to the extent that aspiration occurs on a regular basis which may all contribute to future bacterial colonisation in these patients. This study is a qualitative assessment of the tracheal flora in patients with tracheostomies. The present study demonstrates that tracheostomy is independently associated with lower respiratory colonisation. As seen in the Day 0 culture, lower respiratory tract is sterile, yielding a negative growth in 94.6% cases in our study. Only growth noted in a handful of samples were of *Acenitobacter spps* and *Klebsiella pneumoniae*, but these were isolated only from previously intubated patients who underwent tracheostomy. These patients also were, for majority of the time cared for in the Intensive care unit (ICU) owing to their serious illnesses. Thus, it's note-worthy that external intervention (endotracheal intubation) into the airways of these patients may have led to bacterial colonization. It is also significant to note that most of these patient showed persistent colonization with these organisms even after a week of tracheostomy and even after being on higher antibiotics like Carbapenems. *Acenitobacter spps* and *Klebsiella pneumoniae* are widely regarded as hospital acquired bacteria[10]. Endotracheal intubation predisposes to infections by causing direct effects on airways, reduction of local host defenses, reducing muco-ciliary function, causing stagnation of mucus and increase in entrance of bacteria[11] all of which together predisposes to colonization by these 'hospital acquired bacteriae'. This observation suggests that despite the adherence of strict aseptic precautions, constant exposure to the organisms prevalent in the ICU, results in exogenous acquisition and colonization of the lower airways. A similar picture was noted in previous studies by Jung *et al*[12] and Shanthi M *et al*[13]. It has also to be noted that the fact that most of these patients were on higher antibiotics might have led to development of multi-resistant forms of bacteria, which may partly explain the reason for persistence of colonization. Although strict aseptic precautions are followed, it is without doubt that continued presence of bacteria in the hospital environment does lead to persistent infection. The data in the current study provides further evidence of airway colonization with potentially pathogenic bacteria post-tracheostomy. Patients whose lower airways were colonized post tracheostomy were found to be significantly higher than that noted during the tracheostomy. Our study noted Gram Negative Bacteria (GNB) were cultured more frequently in the samples studied. *Pseudomonas aeruginosa* being the most commonly isolated bacteria. Many other similar studies on tracheostomy have shown that GNB are the most common pathogens causing nosocomial pneumonia[14-16]. In a recent study by Pignatti *et al*[17], in the microbiological analysis performed on tracheal aspirates, *Pseudomonas aeruginosa* was the most predominant bacteria identified. Guimbellot *et al*[18] similarly noted increased development of gram negative bacterial infection predominantly by pseudomonas in children undergoing tracheostomy. Sakurai *et al*[18] studied 15 patients with long term tracheostomies and noted persistent colonization with pseudomonas in them. In another recent study done by Abdollahi *et al*[20], again the predominance of gram negative organisms were noted in a study done on intubated tracheostomised patients but in contrast to our study, majority of the growth was of *Acenitobacter spps* (24%) followed by *Pseudomonas aeruginosa* (16.75%). But this study was done only on intubated patients in contrast to our study in which majority of patients were not intubated prior to tracheostomy. Although the very presence of tracheostomy would have impaired the nasopharyngeal defense mechanisms leading to exogenous bacteria having a direct access to the lower respiratory tract, the more frequent predominance of GNB in the tracheobronchial flora was surprising. This observation suggested that in these, the usual mechanism of airway colonization, which is aspiration of pathogenic GNB from a colonized oropharynx (endogenous route), may not have been the reason for colonisation. Rather, we consider that GNB may have directly entered and become attached to the trachea (exogenous route). Most of our patients were cared for in the ward post-tracheostomy which would have directly lead to exogenous infection. Exogenous route, which is directly into the trachea via the endotracheal tube or via the tracheotomy is one main route suggested for acquisition of infection post-tracheostomy in patients not intubated prior to tracheostomy[5]. There are a lot of factors in play in a hospital environment which may contribute to similar situation. *Pseudomonas aeruginosa* is an opportunistic pathogen but once colonisation is established, it is difficult to be eradicated[21]. Previous classic literatures especially the pioneering work done on the subject by Niedermann[22, 23] has demonstrated that *Pseudomonas spps* can bind more avidly to tracheal cells. It may also have been possible that mechanical injury to the tracheal surface (as could occur from endotracheal intubation, from tracheostomy tubes and suctioning), may expose new binding sites for *Psuedomonas spps*[24]. Another possible explanation being that the reparative changes

which follows this injury allowed for enhanced binding of *Pseudomonas aeruginosa*[25]. Unfortunately, all the studies on bacterial adherence make it clear that the interaction is a complex one, involving multiple surface mucosal and bacterial characteristics. Therefore, it is an uphill task that this process can be interrupted by a single intervention. Perhaps our awareness of micro-environmental factors that influence bacterial interactions will lead to fruitful prevention strategies. It may also be noted that most of these bacteria grown in our culture were just a colonization and did not progress towards a full blown infection. All patients were on antibiotics prior to and after the procedure. Most commonly used antibiotic being the third generation cephalosporin, Cefotaxime. In case of patients in the ICU, antibiotics were chosen by the primary physician. Although we have not studied the antibiotic sensitivity in these samples, we are convinced from previous studies to believe that resistance to cephalosporins in most of the bacteria studied is likely. This deduction is partly due to reason in most of the patients studied, the antibiotic given, Cefotaxime, with a proven action against gram negative bacteria, failed to prevent colonization after one week. In a study by Zakhaur *et al*[16], considerable resistance among GNB of tracheal aspirate was noted in cultures done on tracheal aspirates. Persistent colonization post tracheostomy doesn't also provide a good outlook concerning development of multi-resistant strains causing infection. Various studies have noted the prevalence of GNB in hospitals with multi-resistant forms, a term called healthcare associated pneumonia coined in a manuscript by Restrepo *et al*[26]. Another study[27] by Aliskan *et al* has mentioned the higher risk of potential multidrug-resistant pathogens, including resistant gram-negative organisms, especially *Pseudomonas aeruginosa and Acinetobacter spps*. Recent studies have also implicated the role of bacterial biofilms on tracheostomy tube leading to persistent infection[28], which we do consider, even though we have no evidence from our study, as one of the major cause of persistence colonization in this era of prescribing higher antibiotics and following a strict aseptic care. We do recommend more specific studies are needed on this though. We as clinicians are thus faced with the most difficult question. Whether to start antibiotics prophylactically to prevent infection or to treat infection once it establishes. Either way there are problems and it is indeed a topic of debate even now. According to us, prophylaxis against developing infection is not recommended since this would only facilitate the development of antibiotic resistance in the bacteria, which would make it more difficult to treat the patients once they get infected, but treatment should be purely based on tracheal aspirate culture and sensitivity reports. So, from what we have learnt from our study, we recommend the need for serial tracheal suction tip culture for bacteria and antibiotic sensitivity right from the second or third day post tracheostomy, to study the bacteria, note the development of drug resistance early, so that appropriate treatment can be started as soon as possible thus reducing patient morbidity and mortality. Repeated cultures of bacteria during the course of the treatment thus started would allow for the adjustment of therapy according to changes in flora or antibiotic sensitivity. The same applies to those patients who are intubated, who requires routine endotracheal cultures, owing to the fact that lower airways would be colonized even before tracheostomy is done. During the course of our study, we had over 50 patients for whom tracheostomy was done for prolonged intubation, but they were eventually excluded due to the presence of established lower respiratory tract infection which in majority developed post-intubation. We do also think that, GNB bacteria would have played a prominent role in lower airway infection in these patients. In a study done by Cardenosa *et al*[29] on patients receiving mechanical ventilation, it was noted that most of the patients who were previously intubated and who were on prior antibiotics showed a positive heavy growth of *Acenitobacter spps* and *Pseudomonas aeruginosa* in tracheal aspirate. Also in a recent study by Craven *et al*[30] on 180 patients on mechanically ventilation studied, airways of 22% patients became heavily colonized with a bacterial pathogen and alarmingly, multidrug-resistant pathogens were isolated in 39% of these patients. The presence of a tracheostomy itself, in conjunction with the degree of serious illness seen in patients requiring this procedure, may eventually lead to infectious complications, even causing significant patient mortality. Convincing evidences from our study and others does tend to influence the clinician towards an evidence based therapy but we strongly suggest against it and emphasize the importance of tracheal aspirate culture and sensitivity. Although we were cautious about the role of various host factors which might have influenced the type of bacteria studied, we were convinced from our results that host factors seldom play a role in colonization. We propose further studies on these in the future. We acknowledge the various limitations in our study and possible presence of other confounding factors which may have had a bearing on the final results. Long term studies are required to clearly understand the persistence of colonization and frequency of patients who eventually develop a symptomatic infection following colonization.

CONCLUSION

Colonization of respiratory tract post-tracheostomy is very swift to occur and monitoring of patients with regular tracheal aspirate culture is the most important investigation to identify it. Patients on tracheostomy therapy are at high risk for contracting lower respiratory tract infections which is predominantly due to GNB like *Pseudomonas aeruginosa, Klebsiella pneumonia* and *Acenitobacter spps*. Bacteria like *Acenitobacter spps* were found to be persistently present in previously intubated patients and who were cared for in an ICU. Factors causing colonisation are many, but it is important for us as clinicians to identify this emergence early and treat the patients promptly. This is most important than ever in this era of multi resistant strains of bacteria.

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