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Research Article

Clinical analysis of bacterial flora of lower respiratory tract immediately after tracheostomy and during first tube change: A prospective observational study

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Abstract

Background: Tracheostomy is a deliberate surgical procedure performed to make an opening in the anterior wall of the trachea and maintaining this opening with the use of a tracheostomy tube. Colonization of the tracheobronchial tree with microorganisms almost always follows tracheal intubation, tracheostomy, or the use of ventilatory tubes. Infection of the tracheostomy wound site frequently occurs after prolonged use of the tracheostomy. Objective of the study was to establish the bacteria colonizing the lower respiratory tract in tracheotomized patients. Materials & Methods: We collected 36 endotracheal tube samples very aseptically. The collected specimen was kept in a sterile container and was sent immediately to microbiology department for culture and sensitivity. This was inoculated in thioglycollate broth and incubated for 24 hours at 370 C. After 24 hours the broth was examined primarily for the evidence of growth of the bacteria by direct gram stain smear. Then the sample was swabbed on the antibiotic disc with the sterile cotton swab as per Clinical and Laboratory Standards Institute (CLSI) standard guideline. Results: A total of 36 tracheostomies were performed during the period of this study. There were 29 (80.55%) males with a male: Female ratio of 4.14:1. The age of the patients ranged from 13 months to 78 years. The mean age was 58.08 ± 19.82 years. Patients with upper airway edema from trauma, burns, infection, or anaphylaxis (30.55%) followed by polytrauma and head injury who underwent tracheostomy (19.44%), congenital CNS malformation or disorders (16.66%) and supraglottic or glottic pathologic condition (eg, infection, neoplasm, bilateral vocal cord paralysis (13.88%). Thirty three out of 31 (86.11%) tracheal suction catheter tip cultures yielded a positive result on Day 7 or more. With respect to the identity of the bacteria studied in these positive cultures, they were mainly Acinetobacter baumanni (27.78%), Klebsiella pneumoniae (22.22%), Ps. Aeruginosa (19.44%), Methicillin-resistant Staphylococcus aureus (MRSA) (11.11%) and Acinetobacter/ Pr. Mirabilis/ Candida albicans (8.33% each). Conclusion: This study is a qualitative assessment of the tracheal flora and its antibiotic sensitivity patterns in patients with short term tracheostomies. The present study demonstrates that tracheostomy is independently associated with lower respiratory colonization which subsequently progresses to lower respiratory tract infection.

Keywords: Endotracheal tube aspirates, tracheostomy, culture and sensitivity, gram negative and gram positive bacteria

Introduction

Tracheostomy is an operative procedure that creates a surgical airway in the cervical trachea[1,2]. It is most often performed in patients who have had

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difficulty weaning off a ventilator, followed by those who have suffered trauma or a catastrophic neurologic insult[3].

Infectious and neoplastic processes are less common in diseases that require a surgical airway.

Initially all tracheostomy was carried out only to relieve the upper airway obstruction, gradually its indication became extensive and now it's being increasingly used as temporary procedure for airway access especially for anesthetic purpose and artificial ventilation. Similarly the indication of long term or permanent tracheostomy as in cases of severe respiratory distress, sleep apnoea syndrome and terminal malignant neoplasm are also increasing[4]. General indications for the placement of tracheostomy include acute respiratory failure with the expected need for prolonged mechanical ventilation, failure to wean from mechanical ventilation, upper airway obstruction, difficult airway, and copious secretions[5,6]. The most common indications for tracheostomy are (1) acute respiratory failure and need for prolonged mechanical ventilation (representing two thirds of all cases) and (2) traumatic or catastrophic neurologic insult requiring airway, or mechanical ventilation or both. Upper airway obstruction is a less common indication for tracheostomy[7].

The normal trachea is protected from bacterial colonization, so that the trachea individuals harbors either no bacteria or oral flora in sparse numbers[8]. These defense mechanisms are partially bypassed following a tracheostomy and direct exposure of the lower airways to the pathogens may occur[9]. In case of intubated patients, colonization in the respiratory tract is most common[10].Again, mechanical ventilation is responsible 6 to 10 fold increase the risk of respiratory tract infections[11,12]. In this case tracheal colonization of bacterial isolates may be responsible for added or super infections and at the same time, increases the risk of mortality[13].So, the aim in our study was to detect the spectrum of bacterial isolates and their antibacterial sensitivity in AIMS, Dewas in last one year.

Methods & patients

We collected 36 endotracheal tube samples very aseptically. All the patients who were admitted in ICU of our hospital were on mechanical ventilation. All patients undergoing tracheostomy will be included except those omitted due to exclusion criteria. We collected the data from the enrolled patients in the form of: demographic information, underlying illness, date of admission in our hospital, date of endotracheal tube intubation, date of sample collection and detail of antibiotic therapy prior to collection of samples. The collected specimen was kept in a sterile container and was sent immediately to microbiology department for culture and sensitivity. This was inoculated in thioglycollate broth and incubated for 24 hours at 370 C. After 24 hours the broth was examined primarily for the evidence of growth of the bacteria by direct gram stain smear. Smear was examined in the low power field (LPF) under oil immersion microscope (X100) for detection of squamous epithelial cells and polymorphonuclear nutrophils (PMN). The obtained organism was diluted in 2-3 ml of sterile normal saline. Tube change was done 2 to 11 days after tracheostomy. Tracheostomy tube was removed and new tracheostomy tube was inserted and the endotracheal suctioning was done with sterile suction catheter. In all patients included in the study, after a week of tracheostomy, 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 tip of the suction catheter was cut and kept in sterile container. The tip was sent for bacterial culture and sensitivity.

Then the sample was swabbed on the antibiotic disc with the sterile cotton swab as per Clinical and Laboratory Standards Institute (CLSI) standard guideline[14].Antibiotic disc used from Gram gentamicin, negative bacilli were tobramycin, Netilmicin, amikacin, cefexime, ceftriaxone, ciprofloxacin, ofloxacin, levofloxacin, co-trimoxazole, chloramphenicol, tetracycline, tigicycline, piperacillintazobactum, cefoperazone-sulbactam, ceftazidime. imipenem, meropenem, ertapenem, aztreonam. cefotaxime, polymyxin B, colistin. For Gram positive cases, amoxycillin, oxacilin, amoxycilin-clavauronic acid, piperacillin-tazobactum, cefoperazone-sulbactam, cefuroxime, ceftriaxone, cefexime, ceftazidime, azithromycin, erythromycin, ertapenem, meropenem, imipenem, gentamicin, tobramycin, Netilmicin. amikacin, ciprofloxacin, ofloxacin, levofloxacin, cotrimoxazole, chloramphenicol, teicoplanin, tigicycline, clindamycin, vancomycin, tetracycline, linazolid, polymyxin B, colistin disc were used[13].

Results on continuous measurements are presented on Mean± SD and results on categorical measurements are presented in Number (%). Significance is assessed at 5 % level of significance.

able 1: Demographic and clinical characteristics of tracheotomised patients [n=36]		
Characteristics	No. of the Patients	Percentage [%]
Gender		
Male	29	80.55
Female	7	19.45
Mean Age [Yrs]	58.08 ± 19.82	-
Age Groups [Yrs]		
<20	2	5.55
21-40	7	19.44
41-60	11	30.55
>60	16	44.44
Underlying condition		
Neck trauma	7	19.44
Facial fractures	4	11.11
Cerebral palsy	1	2.77
• Upper airway foreign body	2	5.55
Upper airway anomalies	1	2.77
 Congenital CNS malformation or disorders Supraglottic or glottic pathologic condition 	6	16.66
(eg, infection, neoplasm, bilateral vocal cord paralysis)Cardiac anomaly	5	13.88
• Upper airway edema from trauma, burns,	1	2.77
infection, or anaphylaxis	11	30.55
• Severe sleep apnea	1	2.77
Status of intubation before tracheostomy		
No	3	8.33
Yes	33	91.67

Results

Table 1: Demographic and clinical characteristics of tracheotomised patients [n=36]

A total of 36 tracheostomies were performed during the period of this study. There were 29 (80.55%) males with a male: Female ratio of 4.14:1. The age of the patients ranged from 13 months to 78 years. The mean age was 58.08 ± 19.82 years. Majority of the patients who underwent tracheostomy was >60 yrs (44.44%). Patients with upper airway edema from trauma, burns, infection, or anaphylaxis (30.55%) followed by polytrauma and head injury who underwent tracheostomy (19.44%), congenital CNS malformation or disorders (16.66%) and supraglottic or glottic pathologic condition (eg, infection, neoplasm, bilateral vocal cord paralysis (13.88%). Status of intubation before tracheostomy was positive in 33 (91.67%) cases admitted.

Indication for tracheostomy	No. of the Patients	Percentage [%]
Low GCS	14	38.89
Prolonged ventilation	8	22.22
Respiratory distress	4	11.11
Respiratory Failure		
• Type 1 or Hypoxemic	3	8.33
• Type 2 or Hypercapnic	4	11.11
Type III Respiratory Failure or Perioperative		
respiratory failure	2	5.56
• Type IV Respiratory Failure or Shock	1	2.78

Тε	ıble	2:	Indica	tion	for	tracheostomy	[n=36]

Majority of the patients (38.89%) were for the tracheostomy is low GCS (47.5%) followed by respiratory failure (27.78%) and prolonged ventilation (22.22%) [Table 2].

Table 5: Number of days with endotracheal tube before tracheostomy procedure			
Number of days with ET tube before tracheostomy	No. of the Patients	Percentage [%]	
Not intubated	3	8.33	
Intubated	33	91.67	
1-4 days	5	13.89	
4-7 days	18	50	
>7 days	10	27.78	

Table 3: Number of days with endotracheal tube before tracheostomy procedure

About 50% of the patients in this study were with endotracheal tube for the period of 4-7 days before the tracheostomy. About 27.78 of the patients in this study were with endotracheal tube for the period of >7 days before the tracheostomy [Table 3].

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Bacterial growth	Frequency	Percentage [%]
No growth	33	91.67
Growth	3	8.33
Acinetobacter baumanni	1	2.78
Klebsiella pneumoniae	1	2.78
Ps. Aeruginosa	1	2.78

There out of 36 (8.33%) tracheal suction catheter tip cultures yielded a positive result on Day 0. With respect to the identity of the bacteria studied in With respect to the identity of the bacteria studied in

Bacterial growth	Frequency	Percentage [%]
No growth	5	13.89
Growth	31	86.11
Acinetobacter baumanni	10	27.78
Klebsiella pneumoniae	8	22.22
Ps. Aeruginosa	7	19.44
Staphylococcus	2	5.56
NLFGNB	1	2.78
Citrobacter	2	5.56
Enterobacter	2	5.56
Pr. Vulgaris	2	5.56
Pr. Mirabilis	3	8.33
E Coli (ESBL producer)	1	2.78
Cedecea Lapages	1	2.78
Methicillin-resistant Staphylococcus aureus (MRSA)	4	11.11
Acinetobacter	3	8.33
Mycobacterium chelonae	1	2.78
Mycobacterium fortuitum	0	0
Candida albicans	3	8.33

 Table 5: Bacterial growth pattern in Day 7 Culture [n=36]

Thirty three out of 31 (86.11%) tracheal suction catheter tip cultures yielded a positive result on Day 7 or more. With respect to the identity of the bacteria studied in these positive cultures, they were mainly Acinetobacter baumanni (27.78%), Klebsiella pneumoniae (22.22%), Ps. Aeruginosa (19.44%), Methicillin-resistant Staphylococcus aureus (MRSA) (11.11%) and Acinetobacter/ Pr. Mirabilis/ Candida albicans (8.33% each) [Table 5].

Table 6: Antibiotic sensitivity pattern of the bacteria isolated during tube change					
Sensitivity pattern	Number of patients [n=36]	Percentage [%]			
Penicillin	3	8.33			
Amoxycllin	5	13.89			
Oxacilin	3	8.33			
Amoxycilin-Clavauronic Acid	26	72.22			
Piperacillin-Tazobactum	26	72.22			
Cefoperazone-Sulbactam	23	63.89			
Cefuroxime	9	25			
Ceftriaxone	10	27.78			
Cefexime	9	25			
Ceftazidime	14	38.89			
Azithromycin	18	50			
Erythromycin	11	30.56			
Ertapenem	16	44.44			
Meropenem	26	72.22			
Imipenem	13	36.11			
Gentamicin	11	30.56			
Tobramycin	15	41.67			
Netilmicin	7	19.44			
Amikacin	19	52.78			
Ciprofloxacin	5	13.89			
Ofloxacin	3	8.33			
Levofloxacin	11	30.56			
Co-Trimoxazole	9	25			
Chloramphenicol	13	36.11			
Teicoplanin	24	66.67			
Tigicycline	17	47.22			
Clindamycin	9	25			
Vancomycin	19	52.78			
Tetracycline	2	5.56			
Linazolid	26	72.22			
Polymyxin B	22	61.11			
Colistin	22	61.11			

Table 6: Antibiotic sensitivity pattern of the bacteria isolated during tube change

In our study, incidences of prevalent bacteria were acinetobacter baumannii (27.78%), Klebsiella group (22.22%) pseudomonas aeruginosa (19.44%), staphylococcus (5.56%), E coli and enterobacter group (5.56%). So incidence of acinetobacter was highest followed by Klebsiella. Over all antibiotic sensitivity was observed highly to amoxycilin-clavauronic acid piperacillin-tazobactam (72.22%), (72.22%), meropenem (72.22%), linazolid (72.22%), teicoplanin (66.67%), cefoperazone-sulbactam (63.89%). polymyxin B and colistin (61.11%). Non ESBL and AMPC producer Klebsiella and AMPC producer Klebsiella sensitive to ertapenem, imipenem and meropenem (44.44%, 36.11% and 72.22% respectively), polymyxin B and colistin (61.11%). On the other hand ESBL and AMPC producing Klebsiella were sensitive to ertapenem, imipenem and meropenem (86.11% to 100%) and carbapenemase producing

Klebsiella were highly sensitive to polymyxin B and colistin (75%). Citrobacter were sensitive to chloramphenicol (60%) and polymyxin B and colistin (61.11%) and enterobacter sensitive to polymyxin B (36.11%) and colistin (68.75%) only. Acinetobacter baumannii (both MBL and non MBL producer), pseudomonas aeruginosa (MBL inhibitor) were significantly sensitive to polymyxin B and colistin (80.55%, 80.55 and 86.11% respectively) [Table 6]. **Discussion**

Aspiration of secretions into the lower part of the respiratory tract is a risk factor for pneumonia. [15-18] Many potential pathogens endogenous to the normal oral flora, such as *Staphylococcus aureus* and various species of *Streptococcus*, may be introduced into the lower part of the respiratory tract during intubation[19]. Once a patient is intubated, microaspiration of secretions from above the cuff of the endotracheal tube may occur. Oral secretions can be colonized with endogenous and/or exogenous pathogens. Exogenous pathogens, such as gramnegative bacteria and antibiotic-resistant organisms, can be introduced into a patient's mouth secondary to lack of hand washing and through devices such as oral suctioning equipment. Some organisms, such as *Pseudomonas*, can be transmitted either endogenously or exogenously[19].

Intubation process itself facilitates the entry of bacteria from upper airway into the lower respiratory tract and endotracheal tube further facilitates pooling and leakage of contaminated secretions around the endotracheal cuff. The use of endotracheal & tracheostomy tubes equipped with high volume cuffs and inflation pressure lower than the intra capillary pressure (<30mmHg/<40 cmH₂O) has been proposed to prevent mucosal damage resulting from the pressure exerted by the tube cuff. However narrow longitudinal folds form on the surface of high volume cuff as well as between the cuff and tracheal wall permitting leakage past the cuff. These folds may promote aspiration of regurgitated gastric fluid[20, 21]

In our study total of 36 tracheostomies were performed during the period of this study. There were 29 (80.55%) males with a male: Female ratio of 4.14:1. The age of the patients ranged from 13 months to 78 years. The mean age was 58.08 ± 19.82 years. Saha AK et al¹³ study revealed incidence of positivity in males was 69.17%, which was significant as compared to females (26.25%, p=0.00). In the present study majority of the patients (38.89%) were for the tracheostomy is low GCS (47.5%) followed by respiratory failure (27.78%) and prolonged ventilation (22.22%).

Study by M Hemanth Rao et al study showed that patients with cerebrovascular accidents (CNS) constituted highest percentage (45%) followed by with Polytrauma and head patients injury (22.5%)[22].Respiratory group included 6 patients (15%), of which 3 patients had COPD, 2 patients had cor pulmonale and 1 patient had edema of the upper aero digestive tract. About 38 patients (95%) were intubated before tracheostomy and 2 patients were not intubated before tracheostomy. The most common indication for the tracheostomy is Low GCS (47.5%) followed by prolonged ventilation (27.5%). The third most common indication for the tracheostomy is respiratory failure (15%).

In our study about 50% of the patients in this study were with endotracheal tube for the period of 4-7 days before the tracheostomy. About 27.78 of the patients in this study were with endotracheal tube for the period of >7 days before the tracheostomy. Study

by M Hemanth Rao et al study revealed that first tracheostomy tube change was done between 2 to 9 days (4.10 ± 1.87) [22].Most of the patients underwent first tracheostomy tube change between 3 to 8 days (80%).

In our study, incidences of prevalent bacteria were acinetobacter baumannii (27.78%), Klebsiella group (22.22%) pseudomonas aeruginosa (19.44%), staphylococcus (5.56%), E coli and enterobacter group (5.56%). So incidence of acinetobacter was highest followed by Klebsiella. Study by Ashis Kumar Saha et al¹³ revealed the incidences of prevalent bacteria were acinetobacter baumannii (33.33%), Klebsiella group pseudomonas (31.73%), aeruginosa (18.72%),staphylococcus (2.05%), E coli and enterobacter group (3.65%). So incidence of acinetobacter was highest followed by Klebsiella. Kamath PM et al study noted Gram Negative Bacteria (GNB) were cultured more frequently in the samples studied[23]. 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[24,25].In a recent study by Pignattiet al[14], in the microbiological analysis performed on tracheal aspirates, Pseudomonas aeruginosa was most commonly identified. Guimbellotet al also noted increased development of gram negative bacterial infection in children undergoing tracheostomy[26]. Sakurai et al studied 15 patients with long term tracheostomies and noted persistent colonization with *Pseudomonas* in them [27].

Harlid R et al revealed patients were colonized with one or more potential pathogens at the stomal site and in the trachea in 95% and 83%, respectively, of all sampling occasions [25]. Staphylococcus aureus, gram-negative enteric bacteria (GNEB), and Pseudomonas aeruginosa were the most common colonizing bacteria at these sites. Only 18 of 39 (46%) patients were treated with antibiotics because of RTIs on a total of 30 occasions during the study year.

Present study showed overall antibiotic sensitivity was observed highly to amoxycilinclavauronic acid (72.22%), piperacillin-tazobactam (72.22%), meropenem (72.22%), linazolid (72.22%), teicoplanin (66.67%), cefoperazone-sulbactam (63.89%), polymyxin B and colistin (61.11%). Non ESBL and AMPC producer Klebsiella and AMPC producer Klebsiella sensitive to ertapenem, imipenem and meropenem (44.44%, 36.11% and 72.22%) respectively), polymyxin B and colistin (61.11%). On the other hand ESBL and AMPC producing Klebsiella were sensitive to ertapenem, imipenem and meropenem (86.11% to 100%) and carbapenemase producing Klebsiella were highly sensitive to polymyxin B and colistin (75%). Citrobacter were sensitive to chloramphenicol (60%) and polymyxin B and colistin (61.11%) and enterobacter sensitive to polymyxin B (36.11%) and colistin (68.75%) only. Acinetobacter baumannii (both MBL and non MBL producer), pseudomonas aeruginosa (MBL inhibitor) were significantly sensitive to polymyxin B and colistin (80.55%, 80.55 and 86.11% respectively).

Study by Ashis Kumar Saha et al revealed that ESBL producing Klebsiella pneumonie was highly piperacillin-tazobactam sensitive to (52.63%). polymyxin B and colistin (90.69%)[13]. Non ESBL and AMPC producer Klebsiella and AMPC producer Klebsiella sensitive to ertapenem, imipenem and meropenem (55.81%, 65.11% and 56.97% respectively), polymyxin B and colistin (90.69%). On the other hand ESBL and AMPC producing Klebsiella were sensitive to ertapenem, imipenem and meropenem (90.90% to 100%) and carbapenemase producing Klebsiella were highly sensitive to polymyxin B and colistin (95.65%). Citrobacter were highly sensitive to chloramphenicol (60%) and polymyxin B and colistin (90%) and enterobacter sensitive to polymyxin B (62.5%) and colistin (68.75%) only. Again, gram positive bacteria staphylococcus were highly sensitive to vancomycin, teicoplanin and linazolid (99.99%), chloramphenicol (88.88%) followed by tetracycline and tigicycline (55.55%).

Conclusion

The data in the current study provides further evidence of airway colonization with potentially pathogenic bacteria post-tracheostomy. This study is a qualitative assessment of the tracheal flora and its antibiotic sensitivity patterns in patients with short term tracheostomies. The present study demonstrates that tracheostomy is independently associated with lower respiratory colonization which subsequently progresses to lower respiratory tract infection. 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.

There are several limitations in our study. This was a single center prospective study with flexible inclusion criteria for possible bacterial pneumonia episodes to prevent overlooking possible bacterial infections. Our sample size was not sufficiently large due to the relatively limited number of patients with tracheostomy in limited period of study, and it only reflects experiences from a single medical center. However, the characteristics of respiratory tract infections in patients with tracheostomy can still be deduced from our data.

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