

ORIGINAL ARTICLE

An Epidemiological Study of Bacterial Isolates from Oral Cavity of Medical Students and its Antimicrobial Resistance Pattern

Mohd Saleem¹, S.M.A. Shahid², Mohd Adnan Kausar², Mohammed Qumani Ahmed³, Abdul Rahman Ali⁴, AbdulRahman AlSogair⁴, Abdulaziz Ayed Abed Alrashidi⁴, Abdullah Abdulhadi Hammad Alharbi⁴, Ali Dawood Mohammad Almansour⁴

1- Department of Pathology, Sub-Division Medical Microbiology, College of Medicine, University of Hail, Hail, KSA.
Email: saleemuoh@yahoo.com

2- Department of Biochemistry, College of Medicine, University of Hail, Hail, KSA

3- Department of Pharmacology, College of Medicine, University of Hail, Hail, KSA

4- Student of 4th Year, College of Medicine, University of Hail, Hail, KSA

Abstract

Objective: Isolation and identification of bacteria from the oral cavity of Medical students and teaching staff and determination of antibiotic sensitivity test of the pathogenic isolates.

Methods: From October 2016 to March 2017, a total of 29 specimens were collected from oral cavity. The specimens were taken from medical students. Different media was used for the isolation of the bacterial isolates. Microscopy and biochemical tests were used for identification of bacteria. Kirby Bauer Disc Diffusion method was used for antibiotic sensitivity test of isolated bacteria. **Results:** Mono microbial was seen in 41.4% (12/29) and poly microbial was in 48.6% (17/29). In 29 specimens, 46 bacterial isolates were found. *Streptococcus* species was isolated in 21.7% of bacterial isolates. *Pseudomonas* species and *Staphylococcus aureus* each isolated in 19.6% of bacterial isolates. *Enterococcus faecium* and *Citrobacter* species each isolated in 10.9% of bacterial isolates. The isolation of other organisms was less than 10%. Gram positive isolates was found in 60.9% and Gram negative in 39.1%. Of all the isolates, 97.8% were resistant to Amoxicillin, 84.8% were resistant to Amoxicillin-Clavulanate, 28.3% were resistant to Tetracycline and 8.7% were resistant to Ciprofloxacin. **Conclusion:** In the present study, we were able to isolate and identify several oral bacterial strains which belonged to the species *Streptococcus* species, *Pseudomonas* species and *Staphylococcus aureus* with varying antibiotic resistance patterns.

Keywords: Bacterial isolates, Oral cavity, Antimicrobial resistance pattern

Address for correspondence: Dr. S.M.A. Shahid, Department of Biochemistry, College of Medicine, University of Hail, P.O. Box 2440, Hail, KSA, Mobile:00966-551803367; Email: 2007.sma@gmail.com

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Introduction

Human oral cavity is one of the most dynamic habitats for numerous bacterial species where they undergo intense interspecies competition to form multispecies biofilm structure. Various species of the genus *Streptococcus*, *Lactobacillus*, *Lactococcus*, *Enterococcus*, *Staphylococcus*, *Corynebacterium*, *Veillonella* and *Bacteroids* are the prominent bacteria commonly found in the oral cavity (Rogers, 2008; Wang et al, 2012). Among the oral bacteria, *Streptococcus* and *Enterococcus* are

two important members because they can shift their lifestyle from beneficial microflora on the surface of oral cavity and oropharynx to destructive pathogens when they gain access into the oral tissue and blood stream. Among the diseases caused by oral bacteria include dental caries, periodontitis, endocarditis, pharyngitis, pneumonia, meningitis etc. Most of the oral *Streptococcus* is Gram positive facultative anaerobes demonstrating highly efficient survival strategies such as the ability to adhere hard and soft tissues, cell-cell communication, biofilm formation and to cope up with the

rapidly changing oral environment (Rahman et al, 2015).

The microorganisms found in the human oral cavity have been referred to as the oral microflora, oral microbiota, or more recently as the oral microbiome. The term microbiome was coined by Joshua Lederberg “to signify the ecological community of commensal, symbiotic, and pathogenic microorganisms that literally share our body space and have been all but ignored as determinants of health and disease” (Lederberg and McCray, 2001).

The oral cavity, or mouth, includes several distinct microbial habitats, such as teeth, gingival sulcus, attached gingiva, tongue, cheek, lip, hard palate, and soft palate. Contiguous with the oral cavity are the tonsils, pharynx, esophagus, Eustachian tube, middle ear, trachea, lungs, nasal passages, and sinuses. We define the human oral microbiome as all the microorganisms that are found on or in the human oral cavity and its contiguous extensions (stopping at the distal esophagus), though most of our studies and samples have been obtained from within the oral cavity. Studies have shown that different oral structures and tissues are colonized by distinct microbial communities (Aas et al, 2005, Mager et al, 2003). Approximately 280 bacterial species from the oral cavity have been isolated in culture and formally named. It has been estimated that less than half of the bacterial species present in the oral cavity can be cultivated using anaerobic microbiological methods and that there are likely 500 to 700 common oral species (Paster et al, 2001).

Table-1: Distribution of bacterial isolates

Bacterial isolates	No.	%
<i>Streptococcus species</i>	10	21.7
<i>Corynebacterium species</i>	3	6.5
<i>Neisseria species</i>	3	6.5
<i>Enterococcus faecium</i>	5	10.9
<i>Enterococcus avium</i>	1	2.2
<i>Pseudomonas species</i>	9	19.6
<i>Citrobacter species</i>	5	10.9
<i>Staphylococcus aureus</i>	9	19.6
<i>Klebsiella species</i>	1	2.2

Gram positive isolates was found in 60.9% and Gram negative in 39.1%. Of all the isolates, 97.8% were resistant to Amoxicillin, 84.8%

Materials and Methods

From October 2016 to March 2017, a total of 29 specimens were collected from oral cavity. The specimens were taken from medical students. All the samples/specimens were inoculated on Nutrient Agar and Sheep blood agar media and thereafter incubated at 37°C for 24 hours, for the bacterial isolates (Watt B et al, 1996). Bacterial isolates was identified with the help of Gram staining and biochemical tests as per guidelines of Clinical and Laboratory Standards Institute (CLSI, 2008). Antibiotic sensitivity test was done by the Kirby-Bauer disc diffusion method (Bauer, 1966). Antibiotic Susceptibility/Resistance study for bacterial isolates (CLSI, 2009).

Ethical clearance was taken from Institutional Ethical Committee of College of Medicine, University of Hail, Hail; KSA. The consent was taken from the Medical Students before the collection of specimen.

Results

Mono microbial was seen in 41.4% (12/29) and poly microbial was in 48.6% (17/29). In 29 specimens, 46 bacterial isolates were found.

Streptococcus species was isolated in 21.7% of bacterial isolates. *Pseudomonas species* and *Staphylococcus aureus* each isolated in 19.6% of bacterial isolates. *Enterococcus faecium* and *Citrobacter species* each isolated in 10.9% of bacterial isolates. The isolation of other organisms was less than 10% (Table-1).

were resistant to Amoxicillin-Clavulanate, 28.3% were resistant to Tetracycline and 8.7% were resistant to Ciprofloxacin (Table-2).

Table-2: Distribution of antibiotic susceptibility pattern

Antibiotic#	No. Antibiotic susceptibility pattern					
	Resistant		Intermediate		Sensitive	
	No.	%	No.	%	No.	%
Amoxicillin	45	97.8	1	2.2	0	0.0
Amoxicillin-Clavulanate	39	84.8	5	10.9	2	4.3
Imipenem*	0	0.0	1	5.6	17	94.4
Levofloxacin	2	4.3	13	28.3	31	67.4
Tetracycline	13	28.3	18	39.1	15	32.6
Ciprofloxacin	4	8.7	19	41.3	23	50.0
Vancomycin**	0	0.0	0	0.0	28	100.0

#Multiple response, *In 28 specimens, not tested, **In 18 specimens, not tested

Discussion

Antibiotic resistance raised among commensal bacteria has been supposed to represent a major feature in the development of resistance within bacterial pathogens. In addition, the detection of resistant bacteria in commensal microbiota has pointed to the oral cavity as a possible source for transmission of genes associated to antimicrobial resistance in pathogenic bacteria (Gonçalves et al, 2007).

Nevertheless, studies on the oral cavity and Gram negative bacteria of clinical and epidemiological importance are rare in literature. Despite the limits, these studies are very important for the control of microbial dissemination, and consequently the control of infection rates. The oral cavity can serve as a potential reservoir of *Enterobacteriaceae*, which are spread to the environment and to susceptible individuals through saliva. This fact becomes more important when considering the hospital environment, as most *Enterobacteriaceae* infections take place in different settings (Jorge, 2007; Winn et al, 2012). In this study, *Enterococcus faecium* and *Citrobacter species* each isolated in 10.9% of bacterial isolates.

In the present study, *Pseudomonas species* and *Staphylococcus aureus* each isolated in 19.6% of bacterial isolates. In a study, the colonization rate of the oral cavity by enterobacteria and/or *Pseudomonas species* was 51.0% (Santos and Jorge, 1998).

In the last few years, *Enterobacteriaceae* have been shown to be resistant to a variety of antimicrobial agents. This increase in resistance is primarily related to the frequent use of antimicrobials and to how easy it is for these

microorganisms to build up resistance (Rossi and Andreazzi, 2005; Thomson, 2010).

In the present study, 97.8% were resistant to Amoxicillin, 84.8% were resistant to Amoxicillin-Clavulanate, 28.3% were resistant to Tetracycline and 8.7% were resistant to Ciprofloxacin. In a study (Batabyal, 2012), 73.3% of the isolates were shown to be amoxy/clav resistant *Staphylococcus aureus*.

Batabyal, (2012) demonstrated the development of resistance for amoxycillin/clavulanic by initially; amoxycillin/clavulanic acid was highly effective to treat different Gram positive and Gram negative bacteria. However, after the passage of time, different factors are attributable for emergence of resistance.

Antibiotic resistance is one of the world's most pressing public health problems. The antibiotic resistant organisms can quickly spread and so threaten communities with new strains of infectious disease that are more difficult to cure and more expensive to treat. Treatment failures may arise due to the resistance offered by pathogen against effective broad spectrum antibiotics. These treatment failures and hard to treat infections may results in high death rates (Khushal, 2004).

Conclusion

In the present study, we were able to isolate and identify several oral bacterial strains which belonged to the species *Streptococcus*, *Pseudomonas species* and *Staphylococcus aureus* with varying antibiotic resistance patterns. Above information is also useful to improve healthcare practices and healthcare service, in consonance with the principles of safety.

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