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# Study of Anaerobic Bacteria in Dental Abscess in a Tertiary Care Hospital.

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## ABSTRACT

**Background:** Majority of anaerobes involved in dental infections are thought to be endogenous in origin. Due to breech of continuity of pulp chamber bacterial colonization occurs. Responsible pathogens are polymicrobial. If left untreated in early stages, it can act as foci of disseminated infections and spread rapidly to adjacent structures leading to life threatening conditions. Aims: The present study was undertaken to identify different anaerobic organisms and their association with risk factors. **Methods:** 40 pus samples were collected after mouth wash from patients presented with dental abscess. Samples were processed immediately for aerobic and anaerobic culture. After comparing with the aerobic culture, obligate anaerobes were checked for aero tolerance. Subculture done for identification of species by Gram stain, colony morphology and conventional biochemical tests. Final identification was done by Vitek 2 system. **Results:** 40 (100%) samples were culture positive. Total 60 bacterial isolates recovered from this 40 samples. Out of which aerobes 36 (60%) and anaerobes 24 (40%) isolated. Aerobes present in 18 (45%), anaerobes present in 12 (30%) cases and mixed aerobic and anaerobic flora in 10 (25 %) cases. Predominant isolates were anaerobic cocci, Peptostreptococcus micros (41.6%) followed by Peptostreptococcus anaerobios (25%).Diabetes mellitus, bad chewing habits, poor oral hygiene found as significant risk factors. **Conclusion:** This study highlights polymicrobial nature of infections and role of anaerobes play as pathogens. Early diagnosis and interventions are extremely important to prevent systemic complications. One should have a high index of suspicion of anaerobes while dealing with dental infections.

***Keywords:*** Anaerobes, Dental abscess.

## INTRODUCTION

Dental abscess includes periodontal infections, acute apical abscess and socket infections. Acute dental abscess generally occurs due to necrosis of dental pulp as a consequence of caries or trauma to the tooth or removal of pulp tissue for previous root canal treatment. Abscess formation also occurs due to accumulation of toxic products produced by bacteria which enter the periapical tissues via epical foramen and lead to acute inflammation and pus formation.[1,2]

Anaerobes are most neglected pathogens in dental abscess. Handling of samples containing anaerobic bacteria is very much challenging as they are highly susceptible to environmental oxygen. There are technical difficulties in culture, cost and prolong turnaround time to generate report also discourage dentists to send the sample for anaerobic culture.[3] Majority of anaerobes involved in dental infections thought to be endogenous in origin because they are found as a part of normal flora. Due to breech of continuity of pulp chamber colonization of bacteria occurs. Responsible pathogens are polymicrobial in nature and facultative anaerobes such as viridant group of Streptococci and Streptococcus anginosus group and strict anaerobic cocci, Prevotella spp, Fusobactrerium spp.[4,5]

If the treatment is not initiated in the early stage of the infections then it can act as a foci and rapid spread of infection to the adjacent anatomical structures. Ultimately it may leads to various complications like septicaemia, thrombosis, brain abscess, shock and ultimately death.[6] Most of the localised dental abscesses generally respond to surgical treatment along with adjuvant antimicrobials therapy which prevent spread of the infections.[7] The present study was undertaken from patients presented with dental abscess to identify different anaerobes and their association with various risk factors.

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## MATERIALS AND METHODS

A prospective study was conducted in the Microbiology department of Dr. D. Y. Patil Vidyapeeth, Pune, western India, over a period of 6 months from July 2013 to December 2013. 40 samples were collected from dental abscess patient attending outpatient department of Dr. D. Y. Patil Dental College, Pune. Swab samples from purulent material elicit poor recovery of strict anaerobes. Samples were collected from acute dental abscess patients after mouthwash with hexidine to reduce contamination from normal commensal flora. Then samples were aspirated through intact mucosa or purulent exudates from infected canals by syringe and immediately transported to Microbiology department for processing.[7]

Macroscopic examination of the samples was done. A foul odour, presence of necrotic tissue, were valuable clues for possible presence of anaerobes .Microscopic examination was done for every sample which provides idea about gram positive and gram negative bacteria along with their shape and size. The specimens were inoculated for anaerobic culture into brucella blood agar enriched with vitamin K and hemin, bacteroides bile esculine agar with metronidazole discs (5U) for preliminary identification. All the plates were incubated in anaerobic gaspak jar (BD diagnostics) at 37 0 C and opened after 48-72 hours for inspection of plates. The specimens were also cultured aerobically on 5% sheep blood agar and MacConkey agar & isolates were identified by standard techniques.[8]

Preliminary identification of anaerobic isolates was done by colony morphology, gram stain, aerotolarence test on chocolate agar, fluorescence under long wave (365 nm) ultraviolet light, antibiotic identification discs (Vancomycin 5micro gram, Kanamycin 1000 microgram and colistin 10 microgram), biochemical reactions like catalase test, spot indole test, nitrate reduction test, sugar fermentation tests in viande-Levure broth.[9,10] Automated microbial identification systems, VITEK 2 ANC (Anaerobic and Corynaebacterium) ID card (Bio Mereieux) were used for species level identification. Statistical analysis was performed by Epi Info (TM) 3.5.3.

## RESULTS

Total 40 samples were received for anaerobic culture during the study period in Microbiology laboratory in our tertiary care hospital. All samples were culture positive (100%). Total 60 bacterial isolates recovered from these 40 samples. Out of which 36 (60%) aerobes and 24 (40%) anaerobes which accounting 1.5 isolates per specimen. Aerobes present in 18 (45%) cases, anaerobes in 12 (30%) cases & mixed aerobic & anaerobic flora in 10 (25%) cases [Figure 1].

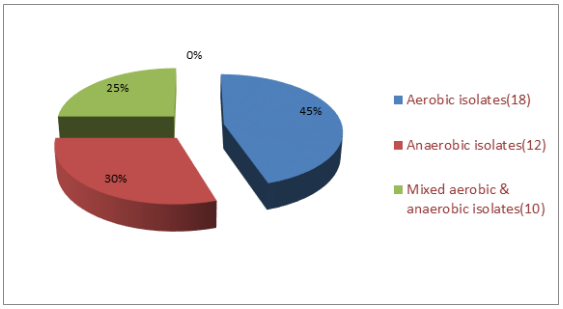


**Figure 1: Distribution of organisms**

Samples were sub grouped into four groups according to the age of the patients. The results from study samples are shown in [Table 1]

**Table 1: Demographic profile**

| Age group (Years) | Number of cases | Number of cases in which anaerobes were isolated | Percentage (%) |
|---|---|---|---|
| 0-20 | 0 | 0 | 0 |
| 21-40 | 11 | 2 | 18.1 |
| 41-60 | 20 | 13 | 65 |
| >60 | 9 | 7 | 77.7 |
| **Gender** | | | |
| Male | 28 | 17 | 60.7 |
| Female | 12 | 5 | 41.6 |

The result of the age base sub group analysis shows an increase incidence in anaerobic isolates with increasing age in cases of dental abscess which is statistically significant. Gender distribution also shown on [Table 1] with no statistical significant value.

Study group also sub divided into three groups according to presence or absence of bad habits like smoking, tobacco or betelnut chewing and good or bad oral hygiene and presence and absence of diabetes mellitus.

**Table 2: Distribution of risk factors**

| Bad habits | Number of cases | Number of cases of anaerobic isolates | Percentage (%) |
|---|---|---|---|
| Present | 28 | 18 | 64.2 |
| Absent | 12 | 4 | 33.3 |
| **Oral hygiene** | | | |
| Good | 6 | 0 | 0 |
| Bad | 34 | 22 | 64.7 |
| Diabetes Mellitus | 12 | 8 | 66.6 |

Above results were analysed by chi square test which gave p value of <0.0001. So the isolation of anaerobes were strongly significant with the history of bad habits and bad oral hygiene and glycemic control.

Various anaerobes were isolated in the study shown in [Figure 2]. In the present study predominant isolates were anaerobic gram positive cocci ie. Peptostreptococcus micros 10 (41.6%) followed by Peptostreptococcus anaerobius6 (25%),

Peptostreptococcus asaccharolyticus 2 (8.3%). Among the gram negative anaerobes Prevotella melaninogenica and Fusobacterium nucleatum isolated in 4 (16.6%) cases.

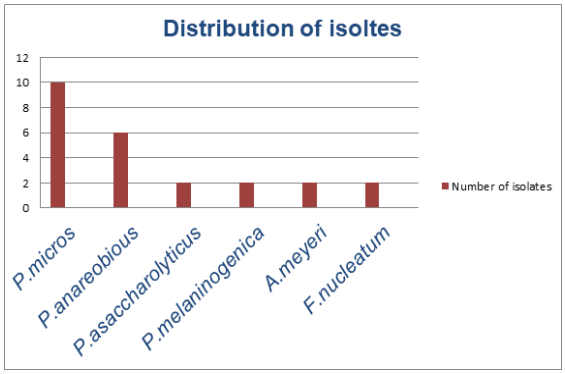


**Figure 2: Distribution of anaerobic bacteria**

In the present study in addition to anaerobic isolates the incidence of aerobic organisms isolated was also studied. [Table 3] showing aerobic isolates in the study group.

**Table 3: Distribution of aerobic isolates**

| Organisms | Number |
|---|---|
| Staphylococcus aureus | 6 |
| Streptococcus viridants | 14 |
| Streptococcus anginosus | 3 |
| CONS | 6 |
| E.coli | 4 |
| Klebsiella pneumoniae | 3 |

## DISCUSSION

Oral infection is one of the most common pathologies among the general population due to infectious complications associated with poor oral hygiene. Prevalence rate of dental infections in India is 65-100%.[11] Dental abscess is one of the common condition if left untreated it can leads to progressive systemic effects along with tooth loss.[12] Predominant treatment can restore microbiota compatible with dental health .Effective therapy implies reduction of pathogenic levels of indigenous oral microorganisms and elimination of exogenous pathogens.[13] For rational treatment and prevention of systemic spread and recurrence we need to identify specific etiological agents.

In our study we have found that isolation rate of anaerobes with increasing age is an important risk factor for dental abscess. Tanaka et al (2002) and Macedo et al. (2006) in their study also observed strong association of dental infections with increasing age.[14,15] Albandar et al (1999) in their study showed that dental infections are higher in male than females which is also similar to our study.[16]

In present study increased anaerobes isolation rate was observed in patients having history of bad habits and association was found to be strongly significant. So, bad habits are important risk factors for dental infections. This may due to increased inflammatory response, decrease resistance caused by these substances and ultimately increasing pathogenicity and number of pathogens in oral cavity. Kent et al (1992) identified bad habits as an important risk factors for dental infections which is similar to our study.[17]

Poor oral hygiene increases the number of pathogenic anaerobes. Present study also showed the statistical significant with the isolation rate of anaerobes among dental abscess patients with bad oral hygiene. Abdellatif and Burt (1987) in their study also showed that oral hygiene is the most important predictor for dental infections in all age groups.[18]

Gram positive cocci, Peptostreptococcus micros (41.6%) and Peptostreptococcus anaerobios (25%) are the predominant organisms isolated in dental abscess in present study followed by Prevotella melaninogenica (9.09%), Actinomyces meyeri (9.09%) and Fusobacterium nucleatum respectively. Few studies have shown varied rate of isolation of gram positive cocci ranging from 2.9% to 43.3% which is also similar to our study.[19,20]

Fusobacterium species and Prevotella species are frequently detected from acute dental abscess. The rate of prevalence found in one study for Fusobacterium species was 73% and other studies suggested rate of isolation of Prevotella species ranging from 10-87% which is differ from our study probably due to small sample size.[21-23]

Facultative anaerobes ie. Viridant group of Streptococci and anginosus group of Streptococci are common in dental abscess and rate of isolation 38.8% and 8.3 %. Staphylococcus aureus and Coagulase negative Staphylococcus (CoNS) isolated in our study 33.3%. In acute dental abscess different study showed rate of isolation for Staphylococcus aureus ranges from 0.7-15 % and in case of CoNS ranges from 4% to 65% which is almost similar to our study.[24-27]

## CONCLUSION

This study highlights that anaerobes play an important role as dental pathogens .Polymicrobial dental abscess and its complications not only a substantial burden on individuals, communities but also in the health care system. Early diagnosis and interventions are extremely important to prevent systemic complications. As most of the anaerobes are sensitive to amoxicillin-clavulenic acid, clindamycin, metronidazole can be used to eradicate the disease. Preventive measures can be taken by improvement in oral hygiene, leaving bad habits and good glycemic control. One should have a high index of suspicion of anaerobes while dealing with dental infections.

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