

Article

Bacteriological profile and their antimicrobial susceptibility pattern in the periodontal lesions

Dr. Pallavi Jain¹, Dr. Parul Nema², Dr. Abhishek Kumar Jain³ and Dr. Ramesh Agrawal^{4*}

¹ MDS Periodontology, Junior Resident, Department of Dentistry, RVRS Govt. Medical College, Bhilwara, Rajasthan.

² Assistant Professor, Department of Pathology, SRVS Medical College, Shivpuri, M.P.

³ Assistant Professor, Department of Microbiology, RVRS Govt. Medical College, Bhilwara, Rajasthan.

⁴ Assistant Professor, Department of Microbiology, NSC Govt. Medical College, Khandwa, M.P.

* Correspondence: drrameshagrwal22@gmail.com

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Abstract: Context: Periodontal disease is a disease, or more likely a number of diseases of the periodontal tissues that results in attachment loss and destruction of alveolar bone. The dental biofilm is responsible for the pathogenesis of the periodontal lesions, resulting both from the direct degradative action and the indirect inflammatory lesions, mediated by microbial cells and their virulence factors.

Aims: The aims of this prospective study were the isolation, identification and characterization of the antibiotic susceptibility profiles of bacterial isolated from patients with periodontal lesions.

Material and Methods: This prospective and non-randomized study was carried out at Department of Periodontology, MPCD & RC, Gwalior and Microbiology, G. R. M. C., Gwalior, Madhya Pradesh for the period of one and half years. The study was conducted for the period of 1 and half year. During the period a total 50 samples were collected following inclusion and exclusion criteria and processed as per Clinical and Laboratory Standards Institute guidelines. The statistical analysis was performed by SPSS software.

Results: A total of 50 study subjects were included, with culture positivity rate of 36% (18). Male to female ratio was 2:1 with mean age of 38.1 years. *E. coli*, *Staphylococcus aureus*, *Streptococcus mutant*, *Klebsiella* species and *Candida* species were isolated. Maximum resistant was observed against ceftriaxone (60%), ceftazidime (53%), amoxicillin clavulanate (46%) and low resistance against gentamicin, ciprofloxacin and doxycycline.

Conclusion: The antibiotic susceptibility assay revealed different profiles and significant levels of antimicrobial resistance, reflecting the necessity to perform the microbiological analysis and the antibiotic susceptibility testing in order to select the optimal antimicrobial therapy for the treatment of the periodontal disease.

Keywords: Periodontitis; Biofilm; Periodontal pathogens.

1. Introduction

Periodontology is the study of the specialized system of hard and soft tissues that supports our teeth and maintains their position in the jaw. The apparatus is known as the “periodontium” [1] or “the attachment apparatus” or “the supporting tissues of the teeth” The periodontium (*peri* = around, *odontos* = tooth) comprises the following tissues: (1) *gingiva* (G), (2) *periodontal ligament* (PL), (3) *root cementum* (RC), and (4) *alveolar bone proper* (ABP) [2]. Periodontal disease is a disease, or more likely a number of diseases of the periodontal tissues that results in attachment loss and destruction of alveolar bone. The natural history of periodontal disease, in some but not all patients, results in tooth loss [3,4]. At present, the prevalence of periodontal disease is approximately 90-95 per cent followed by dental caries affecting nearly 60-80 per cent. Hence, oral problems are to be considered as emerging public health problem in India [5,6].

Periodontitis is caused by a subgingival biofilm consisting of anaerobic and facultative anaerobic bacteria [7].

Patients with chronic periodontitis showed a larger percentage (85%) of gram- negative anaerobic bacteria, like *Aggregatibacter actinomycetemcomitans* a and b serotypes, *Campylobacter rectus*,

Porphyromonasgingivalis, *Prevotella intermedia*, *Tannerellaforsythensis* and *Treponema denticola* [8–10]. Recent studies showed that gram-positive microorganisms, such as *Peptostreptococcus* spp., *Filifactor* spp., *Megasphaera* spp. and *Desulfobulbus* spp. also play an important role in the periodontal disease. *Staphylococcus aureus* strains were isolated in the periodontal pockets of non-smoker patients with aggressive periodontitis [8,11,12].

Treatment of periodontitis involves reduction of the total periodontal bacterial load by supragingival and subgingival mechanical debridement. However, bacterial deposits in the depth of the pockets are often difficult to remove and may be responsible for a poor treatment outcome. Therefore, antibiotic treatment can be indicated for certain patient groups [13]. To provide the patient with an appropriate antibiotic therapy, it is critical to know the susceptibility profiles of clinically relevant oral pathogens [14].

The aim of present study was to identify prevalence of predominantly isolated bacteria and their antimicrobial drug susceptibility patterns among patient with gingivitis and periodontitis.

2. Subjects and methods

The study was conducted at Department of Periodontology, Maharana Pratap College of Dentistry & Research Centre, Gwalior. The study was approved by the Institutional Ethics Committee and a written informed consent was obtained from the all subjects. Subgingival plaque was collected by a single sterile fine paper point inserted until resistance was met into each of the deepest periodontal pockets. After placement for 10 seconds, all paper points were pooled into a vial containing 3-5 ml of sodium thioglycolate broth and transported within 4 to 6 hours at 25°C to the microbiology laboratory for bacterial isolation, identification and antimicrobial susceptibility testing along with requisition form [8,15].

2.1. Procedure Planned

- Patient attending the OPD in Periodontology department.
- Selection of patient under study group on the basis of history and clinical examination after taking consent.
- Specimen collection (subgingival plaque sampling will be taken following strict aseptic precaution into a sterile collection device containing 3-5ml of thioglycolate broth) Figure ??.
- All specimens shall be transported within 4 – 6 hours at 25°C to clinical microbiology laboratory.
- Inoculation of specimen on two sets of Blood agar and MacConkey agar plate.
- One set shall be incubated aerobically and second set incubated anaerobically at 37°C for 24-48 hours.
- Identification of the isolate by culture characteristics, colony morphology, Gram staining, motility and biochemical tests interpretations.
- Antimicrobial susceptibility testing by Kirby Bauer's Disc Diffusion method.

Microsoft Excel® 2010 was used for the compilation of data while SPSS v23.0 and MedCal® v 19.0.5 software were used for statistical treatment. ANOVA test was performed to find out p-value <0.05 (statistically significant at 95% confidence interval.)

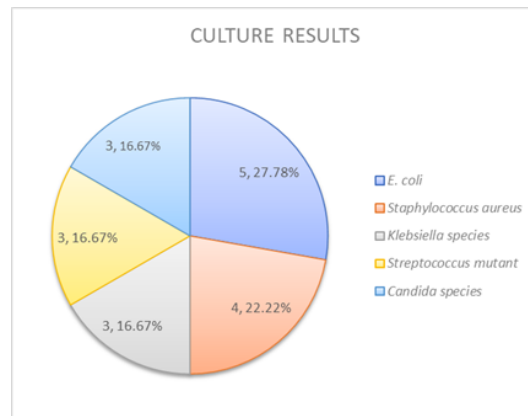
3. Results

A total of 50 patients with periodontitis were recruited into the study. There were 26 males (52 %) and 24 females (48 %). The mean age of the patients was 35.68 years.

Out of the 50 samples assayed via standard culture, only 18 (36 %) were culture positive. Out of them, 3 (16.67 %) were candida species, 8 (44.45 %) gram negative bacilli and 7 (38.95 %) were gram positive cocci. Predominantly isolated organism was *E. coli* 5(27.78%), *Staphylococcus aureus* 4(22.22%) and 3 (16.67%) of each *Klebsiella species*, *Streptococcus mutant* and *Candida species*. Most of the isolates were sensitive to the antibiotics tested. The result showed that 60% of the isolates were resistant to ceftriaxone, followed 53.33% for ceftazidime, 46.67% for combination of amoxicillin and clavulanate; least resistant being 6.67% for gentamicin.

4. Discussion

The discovery of antibiotics has revolutionized the management of infectious diseases more efficiently and timely. As by the famous saying "Every invention and discovery has its own downside", the bacteria are developing resistance to those antibiotics, becoming recalcitrant and troublemaker to treatment [16].



Treatment of periodontitis involves reduction of the total periodontal bacteria load by supragingival and subgingival mechanical debridement. However, bacterial deposits in the depth of the pockets are often difficult to remove and may be responsible for a poor treatment outcome. Therefore, antibiotic treatment can be indicated for certain cases. Antibiotic treatment of periodontitis aims at eradicating or controlling specific pathogens [14,17,18].

In this study, we isolated and investigated the antibiotic susceptibility pattern of the periodontal pathogens from patients with chronic periodontitis of Gwalior region.

Total 50 patients were selected for the study. Age of patients ranged from 20 to 65 years with mean age of 23.56 years. Most of the patients in our study were adults in the age group of 25–50 years as was reported in other previous studies. The probable reason for adults being at higher risk is the higher prevalence of systemic diseases that compromise immunity and the neglect of oral hygiene [19,20].

The male to female ratio presented in our study is 1.08:1, which corresponds with most of the previous studies reported [21–23]. Its probability may be due to the reason that women tends to better oral health and seek oral health care more frequently as compared to male [23].

In our study, aerobic bacterial growth was observed in 36% of the samples. Fungal growth was also observed in 6%. Similarly in 2014, Singh M et al. reported 39% of aerobic bacterial growth [24].

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The microorganisms were divided into two groups as gram positive and gram negative. Gram positive bacteria were observed in 15.21% of the gram stain smear study, negative in 15.21 % of the cases. These findings were partially similar to the findings of Aderhold et al. and Konowet et al. [25,26].

Aerobes were found in following order *E. coli* (10%), *Staphylococcus aureus* (8%), *Candida spp.* (6%), *Klebsiella spp.* (6%) and *Streptococcus mutant* (6%).

In aerobic microbial isolates age group 31-40 years showed highest no. of *E. coli* and *Staphylococcus spp.* with discrete manner.

5. Conclusion

Antimicrobial resistance is a multifaceted complex problem with momentous consequences for individuals as well as health-care systems. The prevalence of multidrug resistance continues to increase among many pathogens, largely because of the overuse and misuse of antimicrobial agents. Such use not only adds to the cost of medical care, but also needlessly exposes the patient to potential toxicity and risks that promote the development and spread of antimicrobial resistance in healthcare facilities. Surveillance of antimicrobial resistance is crucial for providing information on the degree of, and trends in, resistance, and for monitoring the result of interventions. Local observation data are decisive and should be utilized to direct clinical supervision, modernize treatment procedures, instruct prescribers and conduct infection control policies [27].

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