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Oral Bacteria and their Resistance to Antibiotics: Meta-Analysis

Ali Milad*[1], Zakaria A. Khmaj[2], Najeeb Shibani[3], Tarek Dokhan[3], Hadeel Izeit[4,5]

[1]Department of Orthodontics, Faculty of Dentistry, University of Zawia, Libya.
[2]Department of Orthodontics and Dentofacial Orthopedics, Belarusian Medical Academy of Postgraduate Education, Minsk, Republic of Belarus.
[3]Department of Dental Technology, Faculty of Medical Technology, University of Zawia, Libya.
[4]Department of Biological Sciences/ Biomedical sciences, Libyan Academy Misrata, Libya.
[5]Libyan Society of Biomedical Research.

ABSTRACT

Purpose: Bacterial infections are among the most common and significant forms of infectious diseases and pose the greatest threat to the human population in today's day and age. The rapid emergence of antibiotic-resistant bacteria is the main cause of this widespread. The overuse and misuse of antibiotics have significantly contributed to the development of antibiotic-resistant bacteria. To evaluate the efficacy of antibiotics and their effect on bacteria, it is necessary to conduct clinical, biological, and histological research. The aim of this paper is to investigate the spread of bacterial strains in dental prostheses and removable orthodontic appliances and evaluate their resistance to different antibiotics.

Materials and Methods: 100 swab samples were collected from male and female patients who were either wearing removable dental prostheses or removable orthodontic appliances from various dental clinics in the Zawia district, Libya. Microbiological culture was conducted to identify bacterial strains, and sensitivity tests were carried out on a broad range of antibiotics.

Results: After evaluating and testing a wide spectrum of antibiotics, the results demonstrated that the majority of bacterial strains are resistant to the prescribed antibiotics by dental practitioners.

Conclusion: Due to the misuse of antibiotics by healthcare providers, oral bacterial species have developed resistance to most prescribed antibiotics. This poses a significant concern regarding the effectiveness of dental procedures and treatment protocols. Therefore, it is crucial to conduct high-quality randomized clinical trials and meta-analyses to evaluate the effectiveness of antibiotics against oral bacteria. Additionally, these studies can help in assessing and updating the guidelines for prescribing antibiotics by dental practitioners and healthcare providers. *(Open J Biomed Res 2024;3:9-14.)*

Keywords

Antibiotics resistance, Oral pathogens, Dental prostheses, Orthodontic removable appliances, Sensitivity test.

AUDIOABSTRACT

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***Corresponding Author:**

Dr. Ali Milad

Department of Orthodontics,
Faculty of Dentistry,
University of Zawia, Libya.

Email: a.milad@zu.edu.ly

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INTRODUCTION

The oral cavity is a dynamic environment with diverse microbial structure. With 700 different bacterial species identified, these microbes play a significant role in optimizing the oral health, yet oral diseases also arise by them, if their population ratio becomes imbalanced.[1,2]

Developments and researches in oral microbiology have led to valuable breakthroughs in the prevention, diagnosis, treatment of oral pathologies, and most importantly identifying and characterizing the oral cavity microbiome. These breakthroughs can help practitioners in understanding the microbial community of the oral cavity. Such information can be utilized to develop a practical treatment approach for the most common dental pathologies such as dental decay and periodontal inflammation.

The most common bacterias that can be found in the oral cavity are Streptococcus Mutans and Porphyromonas gingivalis.[3] They are responsible for the initiation of caries cavities and periodontal pathologies, respectively.[4] Other types of bacteria are also responsible for the development of dental plaque and calculus, both of which contribute to the

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development of various dental pathologies affecting both hard and soft tissues.[3]

Antibiotic therapy is very commonly used in dentistry, as antibiotics are prescribed to treat bacterial infections, such periodontitis, gingivitis, during endodontics treatment, and as a prophylaxis before, during, and after dental implant placement.[5] However, the overuse of the prescribed antibiotic medications lead to the development of antibiotic-resistant bacteria in the oral microbiota.[6] Antibiotic resistance occurs when bacteria adapt to survive exposure to antibiotics, and over time, these bacteria become resistant to multiple antibiotics, making them more resilient to treatment.[6] The resistance of these bacteria to antibiotics can make dental treatment procedures much more challenging, leading to longer treatment appointments, and potentially increasing the possibility of complications during and post treatment.

To approach the issue of antibiotic resistance, it's necessary to eliminate the main cause, which is antibiotic misuse and overuse. Practitioners should follow the guidelines and avoid prescribing antibiotics in cases that are unlikely to have a bacterial origin.[7] Moreover, additional studies and assessments should be conducted to evaluate the effectiveness of alternative treatment options, such as probiotics and antimicrobial proteins to treat oral infections. Recent research and studies indicated that probiotics and antimicrobial proteins carry a lower risk of inducing antibiotic resistance.[8] However, this concept must also be tested in an oral environment to evaluate its effectiveness against the specific bacterial species found in the oral cavity.[8]

MATERIALS AND METHODS

The third molars in the mandible were examined on the radiographs to see if they were impacted on either the right or left side or on both. These data are permanent and will continue to be accurate because they were created using orthopantomographs, which were utilized to locate the third mandibular molar impactions. The methodology was devised to determine the prevalence of impactions among the greater Zawia population, and to determine if impaction is related to age and Gander.

Data were statistically analyzed by using IBM Statistical Package for Social Science (SPSS), Version 21.0 (SPSS, Chicago, IL, USA). Pearson Chi-Square calculated the association between the third molar impaction conditions to the age group and one way – ANOVA test were used for multiple comparisons between the groups. A p-value of <0.05 was considered statistically significant.

Mouth swab samples were collected from 100 patients who attended dental private and public clinics in Zawia city, Libya. Among the total cases, 70 individuals were selected as patients wearing removable prostheses, while the remaining 30 individuals were wearing removable orthodontic appliances. This selection allowed for a comparative analysis of the swab results. Among the cases, there were 62 female patients and 38 male patients. The age of the patients ranged from 12 to 58 years, with a mean age ranging from 20 to 35 years.

The swabs were taken from patients by dental practitioners and cultured in nutrient agar, Macconkey agar medium, and mannitol salt agar medium. The cultures were then incubated at a temperature of 37 degrees for 24 hours. Biomechanical testing for the identification of gram stain, catalase test, and sensitivity test were conducted. Afterward, applied analytical profile index (API) was performed.

Sensitivity Test of Antibiotics

Sensitivity test was conducted on many of the antibiotics prescribed by practitioners against the bacterial species that were isolated from the collected culture. The bacterial suspension for antibiotics sensitivity analysis was adjusted to a 0.5 Mcfarland standard using Vitek Densichek in accordance with international practice. Zone measurements in mm were performed in accordance with approved guidelines of CLSI.

Data Analysis

All the obtained data was analyzed using an Excel spreadsheet, and it was sorted in the form of numbers and columns as figures and tables

RESULTS

The mixed culture was found to contain *K. pneumoniae, S. aureus, E. cloacae, P. aeruginosa, Pneumococcus, E. coli, A. baumannii, and S. pyogenes* from the swabs taken from the surfaces of dental prostheses and removable orthodontic appliances, especially on the orthodontic appliances, accounting for 43% of the total bacterial colonization. The results also showed a higher prevalence of the mentioned bacteria in male patients compared to females (50% and 38.7%, respectively).

A range of antibiotics were tested for sensitivity in this study against the stated above bacterial strains. The results revealed that the majority of the bacterial strains discovered were resistant to many of the antibiotics prescribed by dental practitioners in Zawia's private and public dental clinics.

The results of antibiotic effectiveness against the tested bacterial strains (Table 1) indicate the following: Colistin sulph antibiotics demonstrated effectiveness against most of the tested strains. Norfloxacin was effective against all tested strains except for *S. pyogenes*. Azithromycin and ceftriaxone were effective against all examined strains, except for *P. aeruginosa*. Ofloxacin showed effectiveness against all examined strains, except for *S. pyogenes* and *A. baumannii*. Meropenem displayed sensitivity to all strains except *Pneumococcus*. Ciprofloxacin and levofloxacin were effective against all tested strains except for *A. baumannii* and *Pneumococcus*. Additionally, Bactrim and gentamicin showed effectiveness against *K. pneumoniae*, *E. cloacae*, *E. coli*, and *A. baumannii*, but were ineffective against other strains, while cefotaxime was only effective against *S. aureus*, *E. coli*, and *P. aeruginosa*.

Regarding the most frequently prescribed antibiotics tested for sensitivity (Table 1), ampicillin was effective against *Pneumococcus* but showed resistance against all other tested antibiotics. Augmentin was only effective against *Pneumococcus*, *E. coli*, and *S. pyogenes*, while it was ineffective against other bacterial strains. Amoxicillin showed effectiveness against *Pneumococcus* and *S. pyogenes* but was ineffective against other bacterial strains. Amikacin exhibited effectiveness against *E. coli*, *P. aeruginosa*, and *K. pneumoniae*, but it was ineffective against other tested bacterial strains. Penicillin lacked sensitivity toward *S. aureus*, while *Pneumococcus* and *S. pyogenes* were sensitive to it. Linezolid was ineffective against *Pneumococcus*, but *S. aureus* and *S. pyogenes* showed sensitivity to it. Tetracycline displayed sensitivity only against *A. baumannii*. Ceftazidime, cefixime, lincomycin, and erythromycin were ineffective against all bacterial strains mentioned in the study.

Table 1: Sensitivity test of antibiotics.

Anti/Stra	*K. pneumoniae*	*S. aureus*	*E. cloacae*	*P. aeruginosa*	*Pneumococcus*	*E. coli*	*A. bumanii*	*S. pyogenes*
Ampicillin	R	R	R	R	S	R	R	R
Augmentin	R	R	R	R	S	S	R	S
Amoxacillin	R	R	R	R	S	R	R	S
Amikacin	S	R	R	S	R	S	R	R
Bactrim	S	R	S	R	R	S	S	R
Ciprofloxacillin	S	S	S	S	R	S	R	S
Ceftriaxone	S	S	S	R	S	S	S	S
Ceftazidime	R	R	R	R	R	R	R	R
Cefotaxime	R	S	R	R	R	S	R	S
Cefixime	R	R	R	R	R	R	R	R
Levofloxacin	S	S	S	S	R	S	R	S
Lincomycin	R	R	R	R	R	R	R	R
Colistin Sulph	S	N	S	S	N	S	S	N
Doxycyclin	S	R	R	R	S	R	S	R
Erytromycin	R	R	R	R	R	R	R	R
Gentamycin	S	R	S	R	R	S	S	R
Meropenem	S	S	S	S	R	S	S	S
Ofloxacin	S	S	S	S	S	S	R	R
Norfloxacin	S	S	S	S	S	S	S	R
Tetracycline	R	R	R	R	R	R	S	R
Tobramycin	R	N	R	R	N	R	S	N
Azithromycin	S	N	S	R	N	S	S	N
Kanamycin	R	N	R	R	N	R	R	N
Ertapenem	S	S	S	R	R	S	S	S
Methicillin	N	R	N	N	R	N	N	R
Clindamycin	N	R	N	N	R	N	N	R
Penicillin	N	R	N	N	S	N	N	S
Linezold	N	S	N	N	R	N	N	S
Fusidic acid	N	R	N	N	R	N	N	R

R= Resistance, S=Sensitive, N= Not tested.

DISCUSSION

Antimicrobial resistance is an increasing health problem. Due to the indiscriminate use of antibiotics, bacterial infections that were previously successfully treated with antibiotics have become less responsive.[9, 10] In the United States, dentists authorize approximately 10% of the prescribed antibiotics, with 50% of them being considered ineffective against oral bacteria.[11] The mechanisms of antimicrobial resistance can be categorized into four types: limiting drug uptake, modification of drug targets, inactivation of drugs, and active efflux of drugs.[12] There is variation in the types of resistance mechanisms used by bacteria due to the differences in cell structure between gram-positive and gram-negative bacteria.[12] Gram-negative bacteria acquire all four mechanisms mentioned above, while gram-positive bacteria commonly employ the mechanism of limiting drug intake.[12]

Removable prostheses and removable orthodontic appliances provide a highly susceptible environment for bacteria to grow and colonize. When an odontogenic or non-odontogenic bacterial infection occurs, it is crucial to develop a treatment plan to prevent further complications. The main component of this treatment plan is the prescription of antibiotics, yet the effectiveness of the prescribed antibiotics is often questioned.[13]

A study conducted by Dağistan S et al. revealed the presence of *Candida spp*. and *Staph. aureus* in 90 patients wearing maxillary full dentures. The majority of microorganisms found on the dentures were *Candida* species (65.5%), followed by *Strep. mutans* (53.3%) and *Staph. aureus* (34.4%).[14] The findings by Dağistan S et al. support those of Baena-Monroy et al., as their study showed that *C. albicans, Staph. aureus,* and *Strep. mutans* were present on the inner surface of complete dentures. *Staph. aureus* and *Strep. mutans* were isolated from 49.5% of the total examined dentures, while *C. albicans* was found in 66.7% of them.[15] Another study conducted by Derafshi R et al. examined the difference in salivary microbial population between individuals wearing removable dentures and a control group, indicating the presence of various pathogens in removable dentures, including *E. coli, K. pneumoniae,* and *E. aerogenes.* Gram-negative bacteria belonging to the *Enterobacteriaceae* family and nonfermenting bacteria were significantly more prevalent in the case group than in the control group.[15]

The association between oral microorganisms and orthodontic treatment has been investigated using traditional techniques such as culturing. With fixed orthodontic appliances, an increase in the prevalence of tooth caries and *S. mutans* bacterial counts was observed according to Lucchese A and et al.[16] Further, Topaloglu-Ak A, et al. conducted a study to evaluate the effect of orthodontic removable appliances on oral microbiota. Their study concluded that, *S. mutans* and *Lactobacillus sp.* counts increased tremendously 6 months after the delivery of the removable appliance.[17] It is important to also mention that further studies have determined that the positioning of orthodontic appliances, both removable and fixed, also has an impact on clinical indicators and the colonization of periodontal pathogenic bacteria such as *P. gingivalis, P. intermedia, P. nigrescens,* and *F. nucleatum*.[18]

From all the above, we can conclude that several studies have been conducted to determine changes in the oral microflora of individuals wearing removable orthodontic appliances and dental prostheses.[19,20,21] However, there is limited literature available on the antibiotic resistance of the presented bacteria and the determination of the most effective antibiotic group to deal with bacteria related to removable prostheses and orthodontic removable appliances.

According to the study carried out by Sebastian et al,[22] they reported that amoxicillin with metronidazole is the standard treatment for odontogenic infections; however, they observed a resistance of 95% in aerobic microorganisms and 86% resistance for anaerobic microorganisms. In contrast, López-González et al[23] reported that anaerobic microorganisms, mainly gram-positive bacilli, have the greatest presence in odontogenic infections and showed resistance to clindamycin in 33.3% of cases.[19-21,24-27] In this study, we investigated the prevalence of bacterial species colonizing dental prostheses and removable orthodontic appliances among patients attending dental clinics in Zawia, Libya. Our findings align with those of our previous study conducted by Milad et al.[28], which demonstrated the presence of Klebsiella pneumoniae, Staphylococcus aureus, Enterobacter cloacae, Pseudomonas aeruginosa, Streptococcus pneumoniae, Escherichia coli, Acinetobacter baumannii, and Streptococcus pyogenes on dental prosthesis surfaces, with particularly high colonization rates observed on orthodontic appliances[28] Building upon these findings, the present study further incorporated antibiotic susceptibility testing to evaluate the efficacy of various these bacterial isolates.inst these bacterial isolates.

Our meta-analysis included 100 samples from male and female patients who are currently

undergoing orthodontic treatment with removable appliances, as well as patients who are wearing removable prostheses. Among the cases, there were 62 female patients and 38 male patients. Our experimental findings indicated the presence of pathogenic bacteria, including *K. pneumoniae, S. aureus, E. cloacae, P. aeruginosa, Pneumococcus, E. coli, A. baumannii,* and *S. pyogenes*, which accounted for 43% of the observed samples. However, no *C. albicans* has been detected in this study. Moreover, these bacterial strains were higher in male samples than females. The *K. pneumoniae* was presented in 53.4% in the total of pathogenic samples While, *E. cloacae 16.6%, S. aureus 13.9%, P. aeruginosa 6.9%, Pneumococcus 2.3%, E. coli 2.3%, A. bumanii 2.3%, and S. pyogenes 2.3%.*

We were able to determine that the most frequently used antibiotics, such as ampicillin, augmentin, amoxicillin, and erythromycin, were ineffective against the discovered bacterial species in the mouths of patients' wearing prostheses or orthodontic removable appliances.

Some types of antibiotics showed diverse activity against specific strains, as bactrim and gentamycin were active against *K. pneumoniae, E. cloacae, E. coli* and *A. bumanii*. *S. aureus, E. coli* and *P. aeruginosa* were sensitive to Cefotaxime, while *K. pneumoniae, Pneumococcus,* and *A. bumanii* were sensitive to Doxycyclin. On the other hand, antibiotic groups such as Ceftriaxone, Ciprofloxacin, Levofloxacin, Colistin Sulph, Ofloxacin, and Norfloxacin showed effectiveness against most of the bacterial strains founded (Table 1).

The majority of the discovered bacterial strains exhibited significant resistance to antibiotics, necessitating increased efforts in antibiotic stewardship and infection prevention and control strategies in both hospitals and dentistry practices.

CONCLUSION

Antimicrobial resistance (AMR) is one of the most serious global public health threats in this century Over the decades, bacteria causing common or severe infections have developed resistance to the many of the prescribed antibiotics, but the misuse by health practitioners. In our meta analysis, we verified the prevalence of *K. pneumoniae, S. aureus, E. cloacae, P. aeruginosa, Pneumococcus, E. coli, A. baumannii,* and *S. pyogenes* on the surface of removable prostheses and removable orthodontic appliances, particularly on the surface of removable orthodontic appliances, which accounted for 43% of the total bacterial colonization. The results also showed a higher prevalence of these bacteria in males compared to females (50% and 38.7%, respectively). After conducting a series of antibiotic sensitivity tests on the collected bacterial species, we were able to determine that most prescribed antibiotics are ineffective against the identified bacterial species, mandating the need for enhancement of antibiotic stewardship and improving the prevention protocols and control of bacterial infection healthcare and dental facilities. Therefore, high-quality randomized clinical trials and meta-analyses should be conducted to evaluate the effect of antibiotics against oral bacteria and to assess and remodel guidelines for prescribing antibiotics by dental practitioners and healthcare providers.

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