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Identification and Antimicrobial Susceptibility of Bacterial Isolates from Odontogenic Abscesses

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Abstract

Objective: The purpose of the study was to identify the bacterial composition of the microbiota from odontogenic abscesses and their antimicrobial susceptibilities.

Study Design: An aspirate of pus from 37 patients with odontogenic abscesses was obtained by needle aspiration and processed aerobically and under anaerobic conditions. Bacteria were isolated and identified by standard Laboratory methods. Then antimicrobial susceptibility of isolated bacteria was determined by using disc diffusion method.

Results: Out of 37 aspirates, 100% yielded positive culture, 34 aspirates contained a mix of microorganisms. A total of 90 strains of bacteria were isolated.Out of 90 strains, 63 strains were anaerobes and 27 strains were aerobes and facultative anaerobes. The mean number of strains per sample was 2.4, two samples were purely anaerobes, 9 samples were mixed anaerobes, 2 samples were purely facultative anaerobes, no purely aerobic, 17 had mixed growth of anaerobes and facultative anaerobes, and 7 samples had mixed growth of aerobes and facultative anaerobic bacteria. Out of 90 isolates, 42 (46.67%) were Gram-positive cocci, 25 (27.78%) were Gram-positive bacilli, 21 (23.33%) were Gram-negative bacilli, and 2 (2.22%) were Candida albicans. The genera of bacteria most frequently isolated were viridans group streptococci, Peptostreptococcus spp., Eubacterium spp., and Prevotella spp. Invitro antibiotic sensitivity of isolated microorganisms were tested for Penicillin, Amoxillin, Ampicillin, Amoxicillin/Clavulanic acid, Erythromycin, and Metronidazole by disc diffusion method. All isolates were sensitive to Amoxillin+ clavulanic acid: 27/27(100%), followed by Ampicillin: 24/27(88.89%), Amoxillin: 23/27(85.19%), Penicillin: 22/27(81.48%), Erythromycin: 12/27(44.44%), and metronidazole: 10/27(37.04%).

Conclusions: The present results confirm the existence of mixed infection with predominance of anaerobic and facultative anaerobic bacteria in odontogenic abscesses. Penicillin still possesses antimicrobial activity against the majority of bacteria isolated from odontogenic infections. However, if penicillin therapy has failed to be effective, the combination of penicillin with ampicillin or amoxillin with clavulanic acid is recommended.

Key words: Odontogenic infections, dental abscess, microbiological diagnosis, antibiotic susceptibility.

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Introduction

Odontogenic infections are among the most common infections of the oral cavity. They can be caused by dental caries, deep restorations that approximate the pulp chamber, pulpitis, periapical abscess, periodontitis, periodontal abscess, and pericoronitis. Odontogenic infections may develop into osteoperiostitis of the jaw, osteomyelitis, and deep fascial space infections. In normal hosts, acute odontogenic infections usually do not occur without some type of predisposing condition, such as periodontal accumulations, necrotic pulp tissue, or tissue damage associated with trauma or surgery [1].

The majority of odontogenic infections is self limiting, and may drain spontaneously. However, these infections may drain into the anatomical spaces adjacent to the oral cavity and spread along the contiguous fascial planes, leading to more severe infection [2]. Due to the proximity of the central nervous system and critical respiratory passages, timely efforts are required to establish a patent airway, mechanical drainage, and appropriate antimicrobial therapy [3].

Odontogenic infections have two major origins: Periapical as a result of pulpal necrosis and subsequent bacterial invasion into the periapical tissue, and periodontal as a result of deep periodontal pocket that allows inoculation of bacteria into the underlying soft tissue [4]. Predisposing factors for odontogenic infections include poor oral hygiene, increased age, uncontrolled diabetes mellitus (especially type 1), neutrophil defects, hormonal changes (e.g., puberty, menstruation, and pregnancy), radiation therapy, chemotherapy, and trauma to the epithelial barrier [5].

Odontogenic infections are usually attributed to the endogenous flora of the mouth, and not to the introduction of non-resident bacteria. An important feature of suppurative odontogenic infections is that they are typically polymicrobial in nature, with mixed aerobic and anaerobic bacteria present [6]. However, the anaerobes generally outnumber the aerobic bacteria by a factor of three to four folds [3, 7, 8].

The predominant bacteria in odontogenic infections such as periapical abscesses or deep fascial space infections are reported to be Fusobacterium nucleatum, pigmented Bacteroides spp., Peptostreptococcus spp., Actinomyces spp., and viridans group streptococci [9] .Even though many patients with odontogenic abscesses show improvement following incision and drainage or tooth extraction, antibiotic therapy can be indicated, especially in acute infections without localized accumulation of pus or in rapidly spreading infections [10,11]. Penicillin (PEN) is the preferred drug in most cases of odontogenic infection, but PEN-resistant organisms have increasingly been isolated from abscesses of odontogenic origin [11, 12]. Therefore, other antibiotics such as clindamycin (CLI), erythromycin, tetracyclines, and levofloxacin (LVX) have been considered as alternative regimens for patients for whom PEN therapy has failed or for patients allergic to PEN [11, 13].

Materials and Methods

This prospective study was conducted at College of Dentistry in Erbil city during period from 1 st August to the end of November 2005. Thirty-seven aspirates of pus from acute odontogenic abscesses were obtained from 37 patients, 26 males and 11 females, age ranging 11-57 years, referred to oral surgery

department in College of Dentistry/ Hawler Medical University in Erbil city.

None of the patients received antimicrobial therapy before specimen collection. Prior to the collection of the specimens, the mucosa had been disinfected with a tincture of povidon-iodin to avoid salivary contamination of the specimen [14].

An aspirate of pus (purulence) from 37 patients with acute odontogenic abscesses was obtained by needle aspiration then inoculated directly in Brain Heart Infusion broth for bacterial activation. After 24 hours of incubation, one plate of Blood agar and a plate of MacConkey agar were seeded for each sample and incubated aerobically and under 5% CO2 at 37 Cº for 24-48 hours. In addition to that all the samples were streaked on other Blood agar plate and Neomycin Blood agar plate, incubated anaerobically using BBL Gas Pack Jar system at 37 Cº for 48-72 hours according to method used by Maza, et al [15].

All bacterial isolates were identified by standard Laboratory Methods [15], then antibiotic sensitivity test (only for aerobic and facultative anaerobic bacteria) on blood and Muller-Hinton agar was done by disk diffusion method against the following antibiotics.

Antibiotics (symbol)	Concentration	Manufactured by
Penicillin (P)	6µg	SDI
Amoxillin (AX)	25µg	OXOID
Ampicillin (AM)	25 µ g	OXOID
Amoxillin/Clavulanic acid (AMC)	20 µg	OXOID
Erythromycin (E)	15µg	OXOID
Metronidazole (MTZ)	5 µg	OXOID

Results

Results of culture finding are represented in (Table 1 and 2). Incidence of isolated anaerobic, aerobic, and facultative anaerobic microorganisms are presented in (Table 3 and 4) respectively. Table (5) gives the results of antibiotic sensitivity test of isolated microorganisms.

Anaerobic bacteria (63 strains within 7 genera) were found in 28 (75.68%) of the 37 specimens. Pure anaerobes only were present in 2(5.41%) specimens, anaerobic/facultative bacteria found in 17 (45.95%) specimens. Two or more anaerobes per specimen were found in 9 (24.32%) of the specimens yielding anaerobes. Pure facultative anaerobic bacteria only in 2(5.41%) and mixed aerobic/facultative anaerobic flora in 7 (18.92%) specimens. Ninety-two percent of abscesses were polymicrobial, with an average of 2.4 isolates per specimen.

The predominant anaerobic bacteria were Peptostreptococcus spp. (20 strains), Eubacterium spp. (14 strains), Prevotella spp. (12), Peptococcus spp. (6 strains), Porphyromonas spp. (5 strains), Actinomyces spp. (4 strains), and Fusobacterium spp. (2 strains). Among the aerobic/facultative isolates from 37 patients with odontogenic infections, 59.26% were Gram-positive cocci, 25.93% were Gram-positive bacilli, 7.41% were Gram-negative bacilli and 7.41% were Candida albicans. The viridans group streptococci were the most abundant

and in most cases associated with anaerobes.

A total of 27 strains of aerobic and facultative anaerobes were tested for their response to six common antibiotics. The percentage of susceptibility for the 27 species were Penicillin: 22/27(81.48%), Amoxillin: 23/27(85.19%), Amoxillin/ clavulanic acid: 27/27(100%), Ampicillin: 24/27(88.89%), Erythromycin: 12/27(44.44%), and metronidazole: 10/27(37.04%).

Discussion

Most acute orofacial infections are of odontogenic origin. Odontogenic infections are typically polymicrobial; however, anaerobes generally outnumber aerobes by at least four fold [3.7,8] . In normal hosts, however, they usually do not occur without some type of predisposing condition. Early recognition and management of acute orofacial infections is critical, because rapid systemic involvement can occur, especially in children. Antimicrobial therapy has an essential role in the management of these infections. If it is initiated before surgery, it can shorten the period of infection and minimize associated risks [2].

In the present study a total of 37 patients with odontogenic infections were examined. Cultivation results of our study indicated that the largest group of isolates is anaerobic bacteria, the most frequently isolated genera were Peptostreptococcus spp. 20 (31.75%), Eubacterium spp. 14 (22.22%), Prevotella spp. (19.05%), and Peptococcus spp. 6 (9.52%) from 37 cases. Regarding to aerobic and facultative anaerobic, viridans group streptococci was the commonest isolates 12(44.44%) followed by Actinobacillus spp. and Lactobacills spp, accounting for 4(14.81%), and 3(11.11%), respectively, from 37 cases.

Our results are in agreement with that of previous studies, in which the number of isolates per specimen ranged from 2.4 to 5 [16, 17, 18]. The mixed aerobic-anaerobic composition of the bacteria involved in suppurative odontogenic infections is thought to be important in the pathogenesis of infection. Barclay reported that if bacteria involved in mixed odontogenic infections are isolated in pure culture and transferred to healthy animals, they are often incapable of producing disease [19]. Thus, a synergistic interdependence between aerobic and anaerobic bacteria is thought to be necessary for the development of infection. Essentially, the respiration of aerobic bacteria depletes the local environment of oxygen, creating an oxygen-poor, nutrient-rich habitat suitable for anaerobic growth. Once anaerobiosis achieved, anaerobes proliferate, secreting toxins and enzymes that result in tissue destruction and abscess formation [20].

The results obtained in the present study, were exceeded the results reported by [21, 22] who found that viridans group streptococci was the predominantly isolated facultative anaerobic species. This also agrees with similar results reported by [22,23,24], while in other studies the greatest prevalence corresponds to gram-negative strict anaerobic bacilli represented by black-pigmented species of the genera Porphyromonans and Prevotella, closely followed by gram-positive strict anaerobic cocci of the genus Peptostreptococcus and gram-negative strict anaerobic bacilli of the genus Fusobacterium [9,25].

The antimicrobial susceptibility results revealed that all the isolates were sensitive to amoxillin+clavulanic acid followed by ampicillin, amoxillin,

penicillin, and erythromycin, but metronidazole shows lowest sensitivity against these bacteria. These results showed that amoxillin+clavulanic acid, ampicillin, and penicillin represent a good choice for treatment of odontogenic infection. This result is consistent with the findings of [23,26,27].

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Table (1): Distribution of No. and percentage of positive culture findings from 37 patients with odontogenic abscesses.

Culture finding	Number of specimens (%)
Total positive culture	37 (100%)
Total positive for aerobic and facultative anaerobic microorganisms	9 (24.32%)
Total positive for anaerobic microrganisms	11 (29.73%)
Total positive for anaerobic and facultative anaerobic microrganisms	17 (45.95%)

Table(2): Analysis of various isolated microorganisms from 37 cases of odontogenic infections.

Microorganisms	**Type of isolate**	**No. (%) of isolates**
Anaerobic and facultative anaerobes	Monomicrobial	2 (5.41%) pure anaerobes
	Polymicrobial	9 (24.32%) mixed anaerobes 17 (45.95%) mixed anaerobes and facultative anaerobes
Aerobic and facultative anaerobes	Monomicrobial	2 (5.41%) pure facultative anaerobes (***Alpha-hemolytic Streptococci)***
	Polymicrobial	7 (18.92%) mixed aerobes and facultative anaerobes

Table (3): Incidence of various anaerobic microorganisms.

Microorganisms isolated	**Number of isolates**	**Percentage (%)**
Gram-positive cocci (26 strains)		
Peptostreptococcus spp	**,18	(31.75%)
Peptococcus spp.	6	(9.52%)
Gram-positive bacilli (18 stains)		
Eubacterium spp.	14	(22.22%)
Actinomyces spp.	4	(6.35%)
Gram-negative bacilli(19 stains)		
Porphyromonas spp.	5	(7.94%)
Prevotella spp.	12	(19.05)
Fusobacterium spp.	2	(3.17%)
Total	63	(100%)

* refers to single bacterial isolate.

Table (4): Incidence of various aerobic and facultative anaerobic microorganisms.

Microorganisms isolated	Number of isolates	Percentage (%)
Gram-positive cocci (16 strains)		
Viridans group streptococci	12	(44.44%)
Group-G B-hemolytic Streptococci	2	(7.41%)
Staphylococcus aureus	2	(7.41%)
Gram-positive bacilli (7 strains)		
Actinobacillus spp	*,3	(14.81%)
Lactobacillus spp.	3	(11.11%)
Gram-negative bacilli (2 strains)		
Klebsiella spp.	2	(7.41%)
Yeast (2 yeast)		
Candida albicans	2	(7.41%)
Total	27	(100%)

* refer to single bacterial isolate.

Table (5): Antibiotic sensitivity test of isolated microorganisms.

N0.	Antibiotic	Concentration of antibiotic disc	N0. (%) of sensitive strains	N0. (%) of resistant strains
1.	Amoxillin	25µg	23 (85.19%)	4(14.81%)
2.	Ampicillin	25µg	24(88.89%)	3(11.11%)
3.	Erythromycin	15µg	12(44.44%)	15(55.56%)
4.	Amoxillin/Clavulanic acid	20 µg	27(100%)	0(00%)
5.	Penicillin	6µg	22(81.48%)	5(18.52%)
6.	Metronidazole	5 µg	10(37.04%)	17(62.96%)