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Original Article

Biochemical Characteristics and Antibiotics Susceptibility of *Streptococcus Mutans* Isolates from Dental Caries in Baghdad City

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ABSTRACT

Background: Genus *Streptococcus* comprises important pathogens that have a severe impact on human health. According to the classification in Bergey's Manual of Determinative Bacteriology, the genus *Streptococcus* includes the pyogenic, oral and anaerobic groups of streptococci, as well as a group of other streptococci. *S. mutans* is a Gram-positive bacterium, which plays a key role in the formation of the dental plaque biofilm as an early coloniser and is the most important bacterium in the formation of dental caries. *S. mutans* do not have flagella, but do have pili and are non-motile facultative anaerobes that grow optimally at 37 °C.

Methods: Eighty samples were collected from patients with different dental caries (pit, fissure and dental roots). The patients had referred to Al-dora Health center and Al-Zewiya Health center in Baghdad city. Bacterial isolates obtained from dental caries samples were streaked on selective medium (Mitis Salivaris Agar) for isolation of *S. mutans*, then the plates were incubated at 37 °C for 48 hours under anaerobic conditions. Bacterial isolates were identified according to their morphological and cultural characteristics and biochemical tests. The disk diffusion method was used to test the antibiotic susceptibility of the selected isolate to different antibiotics. A sterile cotton swab was dipped in fresh culture of *S. mutans* and streaked on the surface of Muller-Hinton agar plates by rotating the plate approximately 60 between streaking to ensure even distribution. The inoculated plates were incubated at room temperature for 10 minutes to allow absorption of excess moisture, then antibiotic disks were fixed by sterile forceps on the surface of plates and incubated at 37 °C for 18 hrs.

Results: Totally, 98 bacterial isolates were obtained. The results indicated that antibiotic susceptibility pattern of the locally isolated *S. mutans* S2 was resistant to two antibiotics (bacitracin and erythromycin), while it was sensitive to the other eight antibiotics, consisting amoxicillin, ampicillin, chloramphenicol, vancomycin, clindamycin, imipenem, gentamycin and cephalothin**.**

Conclusion: Locally isolated *S. mutans* S2 was sensitive to different antibiotics, while it resisted only against bacitracin and erythromycin.

Key words: Biochemical, antibiotic, susceptibility, *Streptococcus mutans*, dental caries.

1. Introduction

Genus *Streptococcus* Rosenbach, 1884 comprises important pathogens that have a severe impact on human health [1]. According to the classification in Bergey's Manual of Determinative Bacteriology, (9th ed.), the genus *Streptococcus* includes the pyogenic, oral and anaerobic groups of streptococci, as well as a group of other streptococci[2]. The cells are spherical or ovoid, 0.5-2.0 µm in diameter, occurring in pairs or chains when grown in liquid media, and stain Gram-positive [2].

The metabolism is fermentative, producing mainly lactate but no gas. The streptococci are catalase-negative, and they commonly attack red blood cells, with either greenish discoloration (α-hemolysis) or complete clearing (α-hemolysis). Optimum temperature for growth is 37 °C, and the growth is usually restricted to 25-45 °C [3,4].

Streptococcus mutans Clarke 1924 is a Gram-positive bacterium, which plays a key role in the formation of the dental plaque biofilm as an early colonizer, producing adhesins which attach the organism to the acquired pellicle of the teeth, and is the most important bacterium in the formation of dental caries. *S. mutans* do not have flagella, but do have pili. On agar. *S. mutans* are Gram-positive ovoid cocci, that typically occur in pairs or chains, are aciduric (grow well in acid medium) and acidogenic (produce acid) and are non-motile facultative anaerobes that grow optimally at 37 °C [5].

S. mutans have a thick cell wall, the cell wall composed of peptidoglycan and teichoic acid that prevent osmotic lysis of cell protoplast, confer rigidity and shape on cell. *S. mutans* have capsule composed of polysaccharide and its structural unit is dextran glucose [6]. The primary habitats for *S. mutans* are mouth, pharynx, and intestine [7]. *S. mutans* metabolize carbohydrates, such as glucose and sucrose, to produce acid and enhance biofilm formation with the early colonizing bacteria to induce dental caries [8].

There is a great interest in the use of antimicrobial agents for prevention and treatment of dental caries due to the spread of antibiotic resistance [9]. Inappropriate antibiotic prescribing and use have been identified as major factors in the emergence of antibiotic resistance in *S. mutans*. Consequently, modification and surveillance of prescribing attitudes have become crucial [10-12]. In the recent years, a shift from narrow-spectrum antibiotic prescriptions which included penicillin to broad-spectrum aminopenicillins with amoxicillin by dental professionals has been reported and the increase of bacterial isolates resistant to the former antibiotics is blamed for such a shift in prescription practices [10,13,14].

The present work was done to isolate and identify *Streptococcus mutans* from dental caries, and study antibiotics, namely, amoxicillin, ampicillin, chloramphenicol, vancomycin, clindamycin, imipenem, gentamycin and cephalothin, susceptibility of *S. mutans* isolates.

2. Materials and methods

2.1. Collection of the sample

Eighty samples were collected from patients with different dental caries (pit, fissure and dental roots). The patients had referred to Al-dora Health center and Al-Zewiya Health center in Baghdad city, Iraq.

2.2. Isolation of *Streptococcus mutans*

Bacterial isolates obtained from dental caries samples were streaked on selective medium (Mitis Salivaris Agar) for isolation of *S. mutans*, then the plates

were incubated at 37 °C for 48 hours under anaerobic conditions.

2.3. Identification of *S. mutans*

Bacterial isolates were identified according to their morphological and cultural characteristics and biochemical tests. Morphological and cultural characteristics including colony size, shape, color, and odor of the bacterial isolates were studied on Mitis Salivaris Agar, the plates after incubation at 37°C for 24 hr. under anaerobic conditions. White biochemical tests included catalase test, blood hemolysis test and carbohydrate fermentation test. All these tests were done according to a previous study [15].

2.4. Biochemical tests

2.4.1. Catalase test [13]

This test was used to detect the ability of bacterial isolates to produce catalase enzyme, which converts hydrogen peroxide to harmLess oxygen and water. Clump of growth from pure culture of each bacterial isolate was transferred onto a microscopical slide using a wooden stick applicator, then two drops of 3% hydrogen peroxide solution were added on bacterial cells. The presence of gaseous bubbles indicates a positive result.

2.4.2. Blood hemolysis test

This test was used to detect the ability of bacterial isolates to produce hemolysin and determine the type of hemolysis, and was achieved by streaking each bacterial isolate on blood agar medium, then incubated at 37 °C for 48 hr. under anaerobic conditions[15].

2.4.3. Carbohydrate fermentation test

This test was used to detect the ability of bacterial isolates to utilize different carbon sources (mannitol, sorbitol and raffinose) as a sole source of carbon and energy. This was achieved by inoculating test tubes containing each carbon source (3%) with each bacterial isolate. All tubes were then incubated at 37 °C for 24 hr. under anaerobic conditions. The change in color of indicator from red to yellow indicates a positive result[16].

2.4.4. Identification by using VITEK-II identification system

The innovative VITEK-II microbial identification system includes an expanded identification database, the most automated platform available, rapid results, improved confidence, and with minimal training time.

Bacterial isolates suspected to be *S. mutans* were completely identified by using VITEK-II identification system. A sterile swab or wooden stick applicator was used to transfer bacterial colonies from a pure culture and was suspended in 3.0 mL of sterile saline in a 12 x 75 mm clear plastic (polystyrene) test tube.

The turbidity was adjusted to 0.8 and measured using a turbidity meter called the DensiChek, then identification cards were inoculated with bacterial suspensions using an integrated vacuum apparatus. A test tube containing cell suspensions was placed into a special rack (cassette) and the identification card was placed in the neighboring slot while inserting the transfer tube into the corresponding suspension tube. The filled cassette was placed either manually or transported automatically into a vacuum chamber station. After the vacuum was applied and air was re-introduced into the station, the cell suspension was forced through the transfer tube into micro-channels that filled all the test wells.

Inoculated cards were passed by a mechanism, which cut off the transfer tube and sealed the card prior to loading into the carousel incubator. All card types were incubated on-line at 35.5, then each

card was removed from the carousel incubator once every 15 minutes, and transported to the optical system for reaction readings, and then returned to the incubator until the next read time. The data were collected at 15-minute intervals during the entire incubation period.

2.5. Antibiotic susceptibility test.

The disk diffusion method was used to test the antibiotic susceptibility of the selected isolate to different antibiotics Table 1. A sterile cotton swab was dipped in fresh culture of *S. mutans* and streaked on the surface of Muller-Hinton agar plates by rotating the plate approximately 60 between streaking to ensure even distribution. The inoculated plates were incubated at room temperature for 10 minutes to allow absorption of excess moisture, then antibiotic disks were fixed by sterile forceps on the surface of plates and incubated at 37 °C for 18 hrs. in an inverted position[17].

Table 1. Antibiotic discs used during this study.

Antibiotic	Abbreviation	Concentration (µg/disk)	Source/origin
Bacitracin	BA	10	**Bioanalyse/Turkey**
Amoxicilin	AmC	10	**Bioanalyse**
Erythromycin	E	15	**Bioanalyse**
Ampicillin	Amp	10	**Bioanalyse**
Chloramphenicol	Chl	30	**Bioanalyse**
Vancomycin	Van	30	**Bioanalyse**
Clindamycin	Cli	20	**Bioanalyse**
Imipenem	IpM	10	**Bioanalyse**
Gentamycin	Gen	10	**Bioanalyse**
Cephalothin	Cph	10	**Bioanalyse**

3. Results

3.1. Collection of samples

In order to isolate *S. mutans*, eighty samples were collected by taking swabs from the mouth cavity of patients suffering from dental caries. These samples were put into peptone water and then streaked on semi-selective Mitis Salivaris agar medium (MSA). Bacterial isolates grown on MSA were then streaked another time on selective Mitis Salivaris Bacitracin agar medium (MSBA). MSBA medium was composed of MSA and 20% sucrose and 0.2 U/mL of Bacitracin, which inhibited the growth of most bacteria excepted *S. mutans* and *S. sorbinus*.

3.2. Identification of bacterial isolates

Bacterial isolates grown on selective medium (MSBA medium) and suspected to *S. mutans* were identified according to their morphological and cultural characteristics and biochemical test.

3.3. Morphological and cultural characteristics

Bacterial isolates grown in Mitis Salivaris Bacitracin agar medium were first identified according to their morphological and cultural characteristics. The results showed that these isolates are gram positive, spherical or ovoid cells. Colonies of these isolates are highly convex, raised, light–blue, frosted glass appearance with either rough or smooth surface. These colonies are also highly adherent to the agar surface if it is picked up by the loop,

polysaccharide parameters were observed as a glistening drop on top of the colony or as a pool besides the colony (Fig 1).

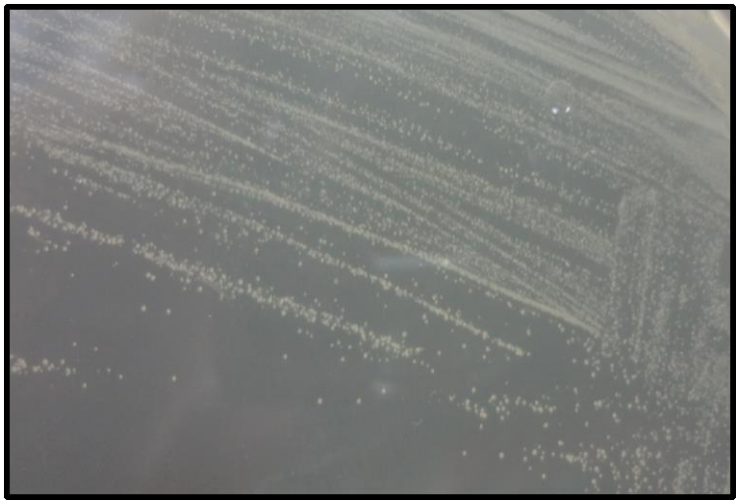

Figure 1. *Streptococcus mutans* on Brain Heart Infusion agar after incubation at 37°C for 16 hours under anaerobic conditions

3.4. Morphological and cultural characteristics

Bacterial isolates grown on Mitis Salivaris Bacitracin agar medium were first identified according to their morphological and cultural characteristics. The results showed that these isolates are gram positive, spherical or ovoid cells. Colonies of these isolates are highly convex, raised, light–blue, frosted glass appearance with either rough or smooth surface as shown in (Fig. 2 and 3). These colonies are also highly adherent to the agar surface and if picked up by loop, polysaccharide formatters was observed as a glistening drop on top of the colony or as a pool besides the colony.

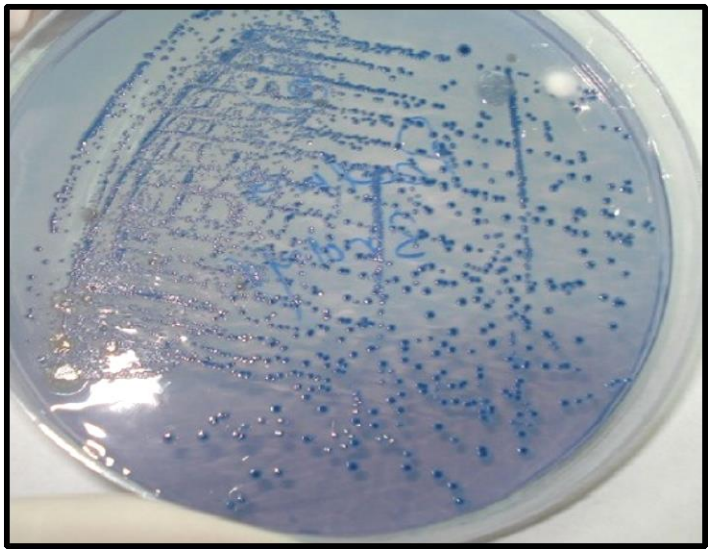

Figure 2. *Streptococcus mutans* on Mitis Salivaris agar after incubation at 37°C for 16 hours under anaerobic conditions

3.5. Biochemical identification

Bacterial isolates suspected to be *Streptococcus* spp. were examined for

their biochemical characteristics. The results showed that these isolates are gram positive, negative for catalase and they were able to ferment mannitol, sorbitol and raffinose sugars (Table 1). These isolates were unable to produce hemolysin exhibiting gamma hemolysis on blood agar medium. According to these results, these isolates were regarded as *S. mutans*.

Table 2. Results of biochemical tests for identification of *S. mutans*.

Biochemical test	Isolate No.									
	S1	S2	S3	S4	S5	S6	S7	S8	S9	**S10**
Gram staining	+	+	+	+	+	+	+	+	+	**+**
Catalase test	-	-	-	-	-	-	-	-	-	**-**
Blood hemolysis	γ	γ	γ	γ	γ	Γ	γ	γ	Γ	**γ**
Acid production from										
Mannitol	+	+	+	+	+	+	+	+	+	**+**
Sorbitol	+	+	+	+	+	+	+	+	+	**+**
Raffinose	+	+	+	+	+	+	+	+	+	**+**
Salt tolerance										
4% NaCl	+	+	+	+	+	+	+	+	+	**+**
6.5% NaCl	-	-	-	-	-	-	-	-	-	**-**

To confirm identification of bacterial isolates as *S. mutans*, biochemical characteristics of these isolates were examined also by using VITEK-Π (Table 3).

Table 3. Results of identification of *S. mutans* according to their biochemical characteristics by using VITEK-Π identification.

Biochemical test	Result									
	S1	S2	S3	S4	S5	S6	S7	S8	S9	**S10**
D-Amygdalin(D-AMY)	+	+	+	+	+	+	+	+	+	**+**
Phosphatidylinositol phospholipase C(PIPLC)	-	-	-	-	-	-	-	-	-	**-**
D-Xylose(Dxyl)	-	-	-	-	-	-	-	-	-	**-**
Arginine dihydrolase1(ADH1)	-	-	-	-	-	-	-	-	-	**-**
Beta-Galactosidase (BGAL)	-	-	-	-	-	-	-	-	-	**-**
Alpha-Glucosidase (AGAL)	+	+	+	+	+	+	+	+	+	**+**
Ala-Phe-Pro Arylamidase (APPA)	-	-	-	-	-	-	-	-	-	**-**
Cyclodextrin (CDEX)	-	-	-	-	-	-	-	-	-	**-**
L-Aspartate Arylamides (AspA)	-	-	-	-	-	-	-	-	-	**-**
Beta-Galactopyranosidase (BGAR)	-	-	-	-	-	-	-	-	-	**-**
Alpha-Mannosidase (AMAN)	-	-	-	-	-	-	-	-	-	**-**
Phosphatase (PHOS)	-	-	-	-	-	-	-	-	-	**-**
Leucine-Arylamides (LeuA)	+	+	+	+	+	+	+	+	+	**+**
L-proline Arylamides (Pro)	-	-	-	-	-	-	-	-	-	**-**
Beta Glucuronidase (BGURr)	-	-	-	-	-	-	-	-	-	**-**
Alpha-Galactosidase (AGAL)	+	+	+	+	+	+	+	+	+	**+**
L-Pyrrdidonyl-Arylamides (PyrA)	-	-	-	-	-	-	-	-	-	**-**
Beta-Glucuronidase (BGUR)	-	-	-	-	-	-	-	-	-	**-**
Alanine Arylamides (AlaA)	+	+	+	+	+	+	+	+	+	**+**

Tyrosine Arylamides (TyrA)	-	-	-	-	-	-	-	-	-	-
D-Sorbitol (dsoR)	+	+	+	+	+	+	+	+	+	+
Urease (URE)	-	-	-	-	-	-	-	-	-	-
Polymyxin B resistance (Poly B)	+	+	+	+	+	+	+	+	+	+
D-Galactose (dGAL)	+	+	+	+	+	+	+	+	+	+
D-Ribose (dRIB)	-	-	-	-	-	-	-	-	-	-
L-lactate alkalinization (lLATk)	-	-	-	-	-	-	-	-	-	-
Lactose (LAC)	+	+	+	+	+	+	+	+	+	+
D-Maltose (Dmal)	+	+	+	+	+	+	+	+	+	+
Bacitracin resistance (BACI)	+	+	+	+	+	+	+	+	+	+
Novobiocin resistance (NOVO)	+	+	+	+	+	+	+	+	+	+
D-Mannitol (d MAN)	+	+	+	+	+	+	+	+	+	+
D-Mannose (d MNE)	+	+	+	+	+	+	+	+	+	+
Growth in 6.5% NaCl (NC 6.5)	-	-	-	-	-	-	-	-	-	-
Methyl-B-D-Glucopyranoside (MBdG)	-	-	-	-	-	-	-	-	-	-
Pullulan (PUL)	-	-	-	-	-	-	-	-	-	-
D-Raffinose (d RAF)	+	+	+	+	+	+	+	+	+	+
O/ 129 resistance (Comp vibrio) (O129R)	-	-	-	-	-	-	-	-	-	-
Salicin (SAL)	+	+	+	+	+	+	+	+	+	+
Saccharose/ Sucrose (SAC)	+	+	+	+	+	+	+	+	+	+
D-Trehalose (d TRE)	+	+	+	+	+	+	+	+	+	+
Arginine dihyrolase2 (ADH2s)	-	-	-	-	-	-	-	-	-	-
Optochin resistance (OPTO)	+	+	+	+	+	+	+	+	+	+

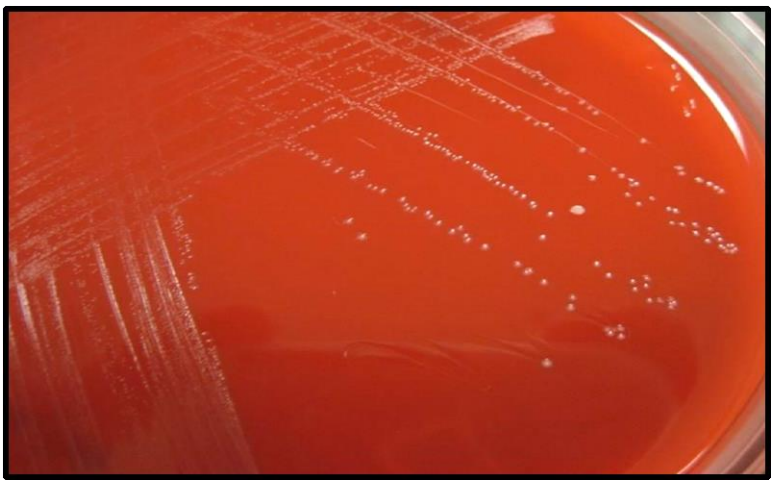

Figure 3. Local isolates of *Streptococcus mutans* on blood agar medium after incubation at 37 °C for 16 hours under anaerobic conditions.

3.6. Antibiotic susceptibility of *Streptococcus mutans*

Antibiotic susceptibility pattern of the locally isolated *S. mutans* S2 against different antibiotics was studied. The results indicated in Table 4 showed that *S. mutans* S2 was resistant to two antibiotics (bacitracin and erythromycin), while it was sensitive to the other eight antibiotics, namely amoxicillin, ampicillin, chloramphenicol, vancomycin, clindamycin, imipenem, gentamycin and cephalothin. The

resistance to these two antibiotics may be encoded by chromosomal DNA and/or plasmid DNA.

Table 4. Antibiotic susceptibility of locally isolated *S. mutans* S2.

Antibiotic	Symbol	Susceptibility
Bacitracin	BA	R
Amoxicillin	AmC	S
Erythromycin	E	R
Ampicillin	Amp	S
Chloramphenicol	Ch	S
Vancomycin	Van	S
Clindamycin	Cli	S
Imipenem	IpM	S
gentamycin	Gen	S
Cephalothin	Cph	S

R: resist S: sensitive

4. Discussion

Bacterial isolates grown on MSA were then streaked another time on selective Mitis Salivaris Bacitracin agar medium (MSBA); MSBA medium was composed of MSA and 20% sucrose and 0.2 U/mL of Bacitracin, which inhibited the growth of most bacteria except for *S. mutans* and *S. sorbinus*. The inclusion of sucrose in this medium led to the formation of glucan and distinctive colony appearance that aided in the identification of *S. mutans* [18]. Bacterial isolates were then streaked on Brain Heart Infusion agar plates and incubated at 37 °C under anaerobic conditions. Colonies of these isolates appeared as points getting into the culture medium and surrounded by a white opaque halo.

According to the classification in Bergey's Manual of Determinative Bacteriology, 9th ed. [19], the genus *Streptococcus* includes the pyogenic, oral and anaerobic groups of streptococci, as well as a group of other streptococci. The cells are spherical or ovoid, 0.5- 2.0 µm in diameter, occurring in pairs or chains when grown in liquid media, and stain Gram-positive. Streptococci require nutritionally rich media for growth. The metabolism is fermentative, producing mainly lactate but no gas. The streptococci are catalase-negative, and they commonly attack red blood cells, with either greenish discoloration (α-hemolysis) or complete clearing (α-hemolysis). Optimum temperature for growth is 37 °C, which is usually restricted to 25-45 °C [3].

Oral streptococci are divided into five different groups: (1) Mutans group (prominent members are *Streptococcus mutans* and *Streptococcus sobrinus*), (2) Salivarius group (*S. salivarius*), (3) Anginosus group (*S. anginosus* and *S. intermedius*), (4) Sanguinis group (*S. sanguinis* and *S. gordonii*), and (5) Mitis group (*S. mitis* and *S. oralis*). Most of the oral streptococci are commensal, nonperiodontho pathogenic bacteria. Some are known to cause infective endocarditis when disseminated through the blood stream, like the early colonizer *S. gordonii*, *S. sanguinis*, and the recently identified *S. oligofermentans* [20]. Oral cavity microbes are often cited as viridans group streptococci because colonies cause greening of blood agar [21].

On agar. *S. mutans* are Gram-positive ovoid cocci, that typically occur in pairs or chains, are aciduric (grow well in acid medium) and acidogenic (produce acid)

and are non-motile facultative anaerobes that grow optimally at 37 °C [5].

It has been inferred that [22] *S. mutans* chromosomal DNA harboring a heterologous erythromycin resistance gene (Erythromycin which is an inhibitor of protein synthesis in susceptible organisms) alters the binding site (ribosome) by methylation the 23s rRNA, thereby, blocks binding of the drug [23, 24]. *Streptococcus mutans* is resistant to bacitracin, which is a peptide antibiotic produced by certain species of *Bacillus*. It has been reported that a gene locus (mbrABCD), which encodes a putative ABC-transporter (MbrAB) and a two-component regulatory system (MbrCD), is involved in the bacitracin resistance of *S. mutans* and that all mutants defective in each mbr gene on this locus are about 100 to120 times more sensitive to bacitracin than the wild-type strain [25]. Al-Mizrakichi (1998) demonstrated that 100% of *S. mutans* isolates showed sensitivity to antibiotic Amoxicillin, gentamycin, Erythromycin, and Cephalothin. It has been demonstrated that *S. mutans* isolates showed sensitivity to Amoxicillin [26]. The results also indicated that *S. mutans* showed sensitivity to Ampicillin. These results are in line with those obtained earlier in the literature[27].

Amoxicillin is routinely prescribed as prophylaxis to the patients prior to massive dental procedures [10]. It has been reported that the introduction of penicillin in the prophylactic treatment has reduced the infection, but the long-term use of penicillin could be compromised by the emergence of resistant strains [28]. Erythromycin and clindamycin have been recommended as alternative options for patients who are allergic to penicillin and are also widely used for antibiotic prophylaxis of endocarditis associated with dental procedures [29]. These two antibiotics have not developed resistance against the three strains of *S. mutans* as revealed by the zone of inhibition that varied between 25mm and 28 mm in the present study. Ofloxacin, doxycycline, tetracycline and chlortetracycline exert their side effects mainly on the digestive system, which include mild stomach pain or upset, nausea, vomiting and diarrhea. They are however effective in inhibiting the growth of *S. mutans* and hence should be recommended for use. Tetracyclines have few side effects but are not recommended for children or pregnant women because they can discolor developing teeth and alter bone growth [30]. Gentamycin, an aminoglycoside, may lead to side effects, which include damage to the ears and kidneys. Metronidazole has been most frequently prescribed by the dental professionals. However, the emergence of resistance to this drug may be slower than if it were used alone, because in order to target both aerobic and anaerobic organisms, metronidazole is used empirically in combination with one or more antibiotics, although the resistance to the drug may be associated with mobile genetic elements, aiding the spread [9].

5. Conclusion

Locally isolated *S. mutans* S2 was sensitive to different antibiotics, while it resisted only to bacitracin and erythromycin. And it was sensitive to the other eight antibiotics, amoxicillin, ampicillin, chloramphenicol, vancomycin, clindamycin, imipenem, gentamycin and cephalothin.

Authors' contributions: Mohssen Hashim Risan and Hameed Majeed Jasim, designed this study, and Aya Raad Salh obtained and analyzed the data. Aya Raad Salh supervised the collection of samples and the identification. Mohssen Hashim Risan, Hameed Majeed Jasim and Aya Raad Salh proceeded to the data quality control and the manuscript drafting.

Mohssen Hashim Risan revised the final version.

Consent for publications

All authors agree to have read the manuscript and authorize the publication of the final version of the manuscript.

Conflict declaration

The authors declare that there is no conflict.

Conflict of interest

None of the authors has any conflict of interest to declare.

Availability of data and material

Data are available on request from the authors.

Funding/Support

None.

Ethics approval and consent to participate

Sample collection were obtained from patients with different dental caries (pit, fissure and dental roots). The patients had referred to Al-dora Health center and Al-Zewiya Health center in Baghdad city.

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References

1. Richards V P, Palmer S R, Pavinski Bitar P D, Qin X, Weinstock G M, Highlander S K, Town C D, Burne R A, Stanhope M J. (2014). Phylogenomics and the dynamic genome evolution of the genus *Streptococcus*. *Genome biology and evolution*, 6(4): 741-753. https://doi.org/10.1093/gbe/evu048
2. Holt G J, Krieg R N, Sneath A H P, Staley T J, Williams T S. (1994). Bergey's Manual Of Determinative Bacteriology. 9th ed: *Williams & Wilkins: Baltimore, Maryland*. 527-558,
3. Grönroos L. (2000). Quantitative and Qualitative Characterization of Mutans *Streptococci* in Saliva and In Dentition. (PhD), Department of Pedodontics and Orthodontics, Institute of Dentistry, University of Helsinki.
4. Salih A R, Jasim H M, Risan M H. (2015). Optimum Conditions for Mutacin Production From *Streptococcus* mutans S2. *Al-Nahrain Journal of Science*, 18(4): 89-97. https://doi.org/10.22401/JNUS.18.4.12
5. Koning J W. (2012). Interactions between *Streptococcus mutans* and *Veillonella dispar*. UCL (University College London).
6. Balakrishnan M, Simmonds R S, Tagg J R. (2000). Dental caries is a preventable infectious disease. *Australian dental journal*, 45(4): 235-245. https://doi.org/10.1111/j.1834-7819.2000.tb00257.x
7. Risan M H, Jasim H M, Salh A R. (2017). Plasmid profile of locally isolated *Streptococcus mutans* in relation with mutacin production. *Iraqi journal of biotechnology*, 16(2): 28-37.
8. Lee S-H, Choi B-K, Kim Y-J. (2012). The cariogenic characters of xylitol-resistant and xylitol-sensitive *Streptococcus mutans* in biofilm formation with *salivary bacteria*. *Archives of oral biology*, 57(6): 697-703. https://doi.org/10.1016/j.archoralbio.2011.12.001
9. Sweeney L C, Dave J, Chambers P A, Heritage J. (2004). Antibiotic resistance in general dental practice—a cause for concern? *Journal of Antimicrobial Chemotherapy*, 53(4): 567-576. https://doi.org/10.1093/jac/dkh137
10. Al-Haroni M, Skaug N. (2007). Incidence of antibiotic prescribing in dental practice in Norway and its

contribution to national consumption. *Journal of Antimicrobial Chemotherapy*, 59(6): 1161-1166. https://doi.org/10.1093/jac/dkm090

11. Rehman F U, Kalsoom M, Adnan M, Fazeli-Nasab B, Naz N, Ilahi H, Ali M F, Ilyas M A, Yousaf G, Toor M D. (2021). Importance of Medicinal Plants in Human and Plant Pathology: A Review. *Int. J. Phar. & Biomedi. Rese*, 8(2): 1-11. http://dx.doi.org/10.18782/2394-3726.1110
12. Khodadadi S, Mahdinezhad N, Fazeli-Nasab B, Heidari M J, Fakheri B, Miri A. (2021). Investigating the Possibility of Green Synthesis of Silver Nanoparticles Using Vaccinium arctostaphlyos Extract and Evaluating Its Antibacterial Properties. *Biomed Res. Int.*, 2021: Article ID: 5572252. https://doi.org/10.1155/2021/5572252
13. Fazeli-Nasab B, Valizadeh M, Hassanzadeh M A, Beigomi M. (2021). Evaluation of the Antimicrobial Activity of Olive and Rosemary Leave Extracts Prepared with Different Solvents Against Antibiotic-Resistant *Escherichia coli*. *Int J Infect*, 8(3): e114498. https://doi.org/10.5812/iji.114498
14. Fazeli-Nasab B, Solouki M, Sobhanizadeh A. (2021). Green Synthesis of Silver Nanoparticles Using an *Ephedra sinica* Herb Extract with Antibacterial Properties. *Journal of Medical Bacteriology*, 10(1,2): 30-47.
15. Collee J G, Miles R, Watt B. (1996). Tests for identification of bacteria. *Mackie and McCartney practical medical microbiology*, 14: 131-149.
16. Yoo S Y, Kim P S, Hwan H K, Lim S H, Kim K W, Choe S J, Min B M, Kook J K. (2005). Identification of non-*mutans streptococci* organisms in dental plaques recovering on mitis-salivarius bacitracin agar medium. *Journal of microbiology*, 43(2): 204-208.
17. Atlas R M, Parks L C, Brown A. (1995). Laboratory manual of experimental microbiology. Mosby-Year Book.
18. Yoo S-Y, Park S-J, Jeong D-K, Kim K-W, Lim S-H, Lee S-H, Choe S-J, Chang Y-H, Park I-S, Kook J-K. (2007). Isolation and characterization of the mutans *streptococci* from the dental plaques in Koreans. *Journal of microbiology*, 45(3): 246-255.
19. Buchanan R, NE G. (1974). Bergey's manual of determinative bacteriology, 7th edn, London. *Bergey Taxon*, 24: 377-378.
20. Kreth J, Merritt J, Qi F. (2009). Bacterial and host interactions of oral *streptococci*. *DNA and cell biology*, 28(8): 397-403. https://doi.org/10.1089/dna.2009.0868
21. Nobbs A H, Lamont R J, Jenkinson H F. (2009). *Streptococcus* adherence and colonization. *Microbiology and Molecular Biology Reviews*, 73(3): 407-450. https://doi.org/10.1128/MMBR.00014-09
22. Li Y, Burne R A. (2001). Regulation of the gtfBC and ftf genes of *Streptococcus mutans* in biofilms in response to pH and carbohydrate. *Microbiology*, 147(10): 2841-2848. https://doi.org/10.1099/00221287-147-10-2841
23. Sartor C, Jacomo V, Duvivier C, Tissot-Dupont H, Sambuc R, Drancourt M. (2000). *Nosocomial Serratia* marcescens infections associated with extrinsic contamination of a liquid nonmedicated soap. *Infection Control & Hospital Epidemiology*, 21(3): 196-199. https://doi.org/10.1086/501743
24. King V M, Bryant S D, Haque M N. (2001). Fact and Fantasy Regarding Resistance to Microbicide. *Journal of the American Leather Chemists Association*, 96(5): 162-168.

25. Tsuda H, Yamashita Y, Shibata Y, Nakano Y, Koga T. (2002). Genes involved in bacitracin resistance in *Streptococcus mutans*. *Antimicrobial agents and chemotherapy*, 46(12): 3756-3764. https://doi.org/10.1128/AAC.46.12.3756-3764.2002
26. Jain P, Pundir R K. (2009). Antibiotic sensitivity pattern of *Streptococcus mutans* against commercially available drugs. *Journal of Pharmacy Research*, 2(7): 1250-1252.
27. Al-Mizrakchi A. (1998). Adherence of mutans *Streptococci* on teeth surfaces: microbiological and biochemical studies. PhD Thesis.
28. Fani M, Kohanteb J, Dayaghi M. (2007). Inhibitory activity of garlic (*Allium sativum*) extract on multidrug-resistant *Streptococcus mutans*. *Journal of Indian Society of Pedodontics and Preventive Dentistry*, 25(4): 164-172.
29. Bagg J, MacFarlane T W, Poxton I R, Smith A J. (2006). Essentials of microbiology for dental students: *Oxford university press*.
30. Roberts M C. (2002). Antibiotic toxicity, interactions and resistance development. *Periodontology 2000*, 28(1): 280-297.

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