

Isolation Gram Positive Bacteria from Deep Carious Lesion Iraq-Baghdad.

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Abstract

Streptococcus mutans is one of cariogenic microorganisms associated with tooth decay. The ecological plaque according with the hypothesis and caries is the changes consequence in the natural dental plaque microflora. 100 specimens were according to be positive bacterial isolates by using Mitis-Salivarius agar medium. 98% for total isolates were considered *Streptococcus* and specially *S. mutans* identification depending on biochemical test and Vitech 2 system.

The common species strain according to (eight strains) including *Bacillus* sp., *Staphylococcus* sp., *Enterococcus*, *Faecalis*, *Streptococcus* sp, aerobic bacteria and *Lactobacillus fermentum*,

Keywords: human dental plaque, streptococci bacteria, teeth caries, periodontal.

1. Introduction

In human Oral cavity supports life for different variety species bacteria permeate into the tissues same time lower host immunity. Loesche [1] different types reach about 700 of species include groups bacteria that found in the mouth, it is important to healthy oral preserving. Growth comprising lactic acid bacteria with mouth gives an environment genera *Lactobacilli*, *Streptococcus*, found in mouth and droplet spray *Entamoeba gingivalis* [2].

A Necessary Factor of Etiology Dental Diseases Bacterial of Infection Both Dental Caries and Periodontal Diseases ,Still affect a Major Portion of the Population [3].

These findings make the control of oral microbes a vital concern because dental plaque biofilm protects pathogenic bacteria from antibiotics. Increasing resistance among pathogens to conventional antibiotics and side effects of current therapies have made plants traditional medicine a potential source to screen for antimicrobial agents Firmino et al. [4] Were formerly Microbes with oral diseases have been shown ingeneral pathogenic. Almost 50% microflora oral streptococci., but tooth brushing vigorous after Bacteremia may occur dental treatment after in patients with periodontitis Thus, for many species ,oral cavity acts as an important pathway into the human body [5].

The development of caries bacteria is considered as the pioneer in, in dentin especially, and have an antagonistic effect against pathogens periodontal. The inhibition of their growth is through the production of acids which lowers the environment's pH release bacteriocins and (H₂O₂), playing a key role in maintaining the balance of antimicrobials in the mouth and the digestive system Al-hammar et al. [6] Generally Periodontal disease include alveolar ,gingival, bone, and cementum to pathologic state inflammatory of the gingiva and the supporting structures of the periodontium. Epidemiological data in United States, that affects one-half periodontal disease of its population is the important cause of tooth loss among adults [7].

This goal research isolation and identification of microorganisms causes teeth decay.

2. Materials and Methods

Samples of Collection

100 samples collected from 100 patient dental carries. who had refrained from eating, drinking and oral hygiene for 2 hours before collection in AL Yarmouk hospital, Baghdad, in sterile transport media then transport in cool box to the laboratory.

A/ Isolation procedures

A stained with gram stain by doing smears and to bacteria isolated. By using light microscope [8].

B/ used culture

Spacemen was incubated into both (Blood and Mitis Salivarius); incubated under anaerobic conditions. differentiate between strain by Biochemical reactions [9].

Bacterial Isolates

Samples were streaked on selective Mitis Salivarius Agar to obtain isolated colonies of oral streptococci species. Samples were grown under anaerobic conditions (jar under CO₂ at 37°C for 24 hrs). Colonies demonstrating dissimilar colonial morphology with gram positive cocci arranged as chains were chosen in the present work and MRS medium.

For isolation *Lactobacillus* while Esculin Agar for *Enterococcus* Sp. Confirmation identification optochin sensitivity and by Vitech 2 system. [10].

3. Results and Discussion

Isolation of Bacterial Isolates

In gram positive the type of bacteria isolated belong to, Table (1) the isolated were the aerobic bacteria eight strains according *Streptococcus* spp, *Enterococcus* spp, *Bacillus* sp., *Staphylococcus* sp., anaerobic bacteria which include *Lactobacillus fermentum*,

Table 1: Number and type of bacteria isolated from teeth caries

Type of microorganism (Grams positive)	Number of microorganism (100%)
<i>Streptococcus mutans</i>	37

Streptococcus salivarius	33
Streptococcus. Sanguis	28
Enterococcus. Faecalis	25
Staphylococcus aureus	20
Bacillus cereus	16
Staphylococcus epidermidis	17
Lactobacillus fermentum	10

All samples collected were streaked on Mitis Salivarius Agar; a selective medium used to isolate oral streptococci and differential because each species of streptococcus has a distinguished colony morphology. The medium contains selective that bactericidal Gve+ bacilli except streptococci and enterococci. (Potassium tellurite) salt is reduced by Enterococcus Spp. Form black colonies. *S. salivarius* sucrose metabolizes and colonies develops with a "gum-drop" appearance. *S. mitis* and enterococci do not metabolize sucrose and, form smaller colonies on this medium. Colony morphology of Streptococcus species obtained on Mitis salivarius agar, as seen under a microscope (Figure 1) and blood agar colonies Single on the surface of MS-agar by subculturing and purification on blood agar. Gve + included cocci which are (spherical or ovoid) chains under light microscope

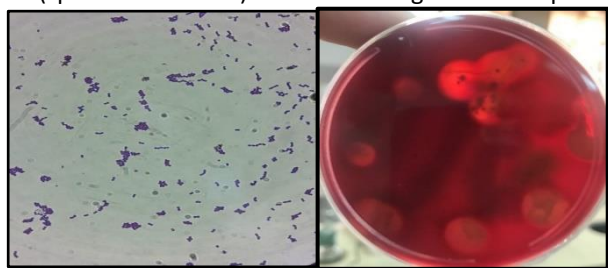


Figure (1) A: Gram staining–*S. mutans*, B: Blood agar at 37c for 24 hrs

Streptococcus mutans was the highest counts was obtained from carries samples followed by complete denture group (Figure 2). The *S. mutans* levels was very low decreasing by dentulous, removable partial denture and edentulous groups. Levels of *S. sanguis* were similar to that *S. mutans* except for slight predominance in the complete denture group. The result agree with Mohapatra et al. [3]. The *Enterococcus* spp was second bacteria predominate dental caries see (Figure 3).



Fig (2): Colony counts of A) *S. mutans* B) *S. sanguis* on Mitis Salivarius Agar at 37c anaerobic jar for 24hrs

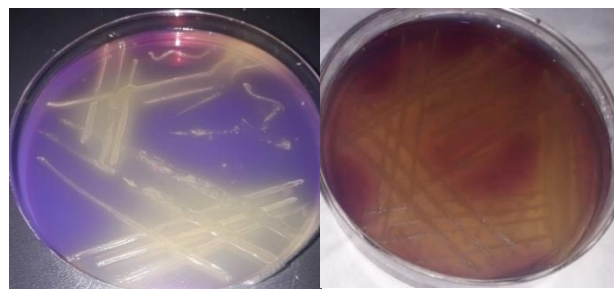


Fig (3): Colony *Enterococcus* spp. on Esculin Agar After 24 hr.at 37°C

generally considered and associated with systemic diseases several, ental caries, tooth decay commonly known as, oral health problem is the most common and *S. mutans* is considered to be the primary agents of dental caries [11, 12].

Identified depending of isolates on the surface of MS-agar media. colonies raised that were identified as *S. mutans* which was considered as one of the important agent of etiological dental caries [13] by Asokan et al. [14]

Result disagree with Mohapatra et al. [3] shoven patients about reached 30 cases have *s. mutans* and another *Staphylococcus* spp, that main higher than *s. mutans* .

A positive bacteria reached (10^4 cells/ml) colonies on MS-agar were selected by sub culturing [12].

An imbalance between demineralisation and remineralization dental caries occurs because (demineralization) is higher than (remineralisation), caries can occur [15].

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