INT. J. BIOL. BIOTECH., 20 (1): 03-16, 2023.

VIRIDANS GROUP STREPTOCOCCI AND DENTAL CARIES: AN OVERVIEW

Nazia Masood Ahmed, Perween Tariq and Asma Naim

Department of Microbiology, University of Karachi, Karachi-75270, Pakistan
email: nazia.masood@gmail.com

ABSTRACT

Worldwide, dental caries is a multifactor prevalent disease, more widely known as tooth decay which ultimately results in loss of tooth and will continue to develop and progress for an individual's entire life if not paid attention to. Viridans group streptococci (VGS), facultative normal flora of oral cavity are the major etiologic agents of dental caries (oral infection). It is also a common and serious problem in Pakistan which is found to be approximately 30% to 60%. Hardly, few individuals understand the true implication of dental caries and what measures can be used to stop it.
VGS have been thought to be of low virulence because they are not primary pathogens. The cariogenicity of oral streptococci is attributed to their acidogenic potential and glucan (extracellular polysaccharide) production. However, VGS can act as opportunistic pathogens under favorable conditions and also responsible for causing extra-oral infections such as infective endocarditis, bacteremia and pneumonia. The present study reviews the general characteristics of VGS and preventative measures for dental caries.

Keywords: Viridans group streptococci, glucan, dental caries, endocarditis, tooth loss.

INTRODUCTION

ORAL MICROBIAL FLORA

The human body is connected to the external environment through a number of body openings or sites such as the gastrointestinal tract with two openings; the mouth and the anus, the urogenital opening and the nasal passage. All these body openings are inhabited by diverse transient and resident microbial communities which contribute towards the first line of defense against pathogenic organisms (Ferrer *et al*., 2021; Brooks *et al*., 2002).

The oral cavity forms the first portion of the alimentary canal. The mouth is normally kept moist due to the continuous production of saliva. It is lined with a mucus membrane. Moreover, the health of teeth and gums influences the overall health of the body. The oral cavity harbors a complex population of microorganisms. A normal mouth is colonized mostly by commensal bacteria, part of these organisms are restricted to the oral cavity. The microbial flora are complex in nature and approximately more than 800 distinct species are reported as normal inhabitant of oral cavity (Ferrer *et al*., 2021). The primary colonizers of the teeth surface are mainly streptococci. Among streptococci, the major species are *Streptococcus salivarius*, *S. mutans*, *S.oralis*, *S. mitis*, *S. sanguinis* and *S. gordonii*. Along with the *Streptococcus* species, different other genera are also present in the oral cavity such as *Actinomyces*, *Veillonella*, *Gemella*, *Staphylococcus*, *Lactobacillus*, *Haemophilus*, *Bacteroides*, *Fusobacterium*, *Abiotrophia* and *Granulicatella* (Elavarasu *et al*., 2012)

FACTORS CONTRIBUTING TO MICROBIAL DIVERSITY

The diversity of microbial community can be attributed to the complex environmental conditions prevailing in the oral cavity i.e., pH, temperature, oxidation reduction potential, composition of saliva, dietary habits and availability of minerals, salts and other nutrients (Bjorklund *et al*., 2011; Kuribayashi *et al*., 2012). The flora differs from one locus to other because of diversity in their nature and due to variation in the environmental factors, variation in origin, and having different metabolic status (Chattoraj *et al*., 2011). The regulatory forces influencing the oral microbiota can be classified into three main categories i.e., host related, microbe related and external factors (Marcotte and Lavoie, 1998).

VIRIDANS GROUP STREPTOCOCCI

Viridans Group Streptococci (VGS) is the most prominent group of bacteria found in the oral cavity (Ergin, 2010; Maeda *et al*., 2011). VGS are heterogeneous facultative anaerobes, residing as commensal flora in the oral cavity, gastrointestinal tract, respiratory tract, urogenital tract, skin, nasal cavities and the oropharynx (Ono *et al*., 2000; Hillman *et al*., 2009). Under favorable conditions, VGS can act as opportunistic pathogens, involve in number of oral and extra oral infections and the most common are dental caries, endocarditis, meningitis and pneumonia especially in immunocompromised/neutropenic patients (Brook, 1994; Tunkel and Sepkowitz, 2002 and Liu *et al*., 2012). VGS can also produce infections in mutual relationship with other microbial flora (Law *et al*., 2007).

CLINICAL CONDITIONS ASSOCIATED WITH VGS

Clinical symptoms of VGS include both oral and extra-oral infections. The most common oral infections are dental caries, periodontal disorders, gingivitis, perioral abscess, recurrent aphthous stomatitis, dentoalveolar and suppurativeorofacial infections (Hardie, 1992; Blandino*et al*., 2007; Belda-Ferre*et al*., 2012; Martinez-Martinez *et al*., 2019 and Lam *et al*., 2022).

Besides oral infections, VGS are also responsible to cause different extra-oral infections such as endocarditis, congenital heart disease, myocardial and cerebral infarction (Shelburne *et al*., 2007), pneumonia, acute bronchopulmonary infections, empyema thoracis and lung abscess, sepsis (Presterl *et al*., 2005), nosocomial infections (Lyytikainen *et al*., 2004), liver abscess, bacteremia (Ergin, 2010), acute and chronic urethritis (Fang *et al*., 2012), fatal shock syndrome (Steiner *et al*., 1993), Reiter syndrome (Huang *et al*., 2000), meningitis, brain abscess (Harrell and Hammes, 2012), cavernous sinus thrombosis (Chang *et al*., 2003) and septic arthritis (Martinez-Martinez *et al*., 2019; Edson *et al*., 2002).

Among these extra-oral infections caused by VGS, endocarditis is by far the most prevalent infection. VGS enter the blood stream as a result of poor oral hygiene, presence of some dental disease or dental manipulation involving gingival margin and cause transient bacteremia. The bacteremia could be responsible to cause endocarditits particularly if heart valves are damaged by previous rheumatic fever or by cyanotic heart disease (Brook *et al*., 2002).

CLASSIFICATION OF VGS

The taxonomic classification of the VGS has been under continuous evolution due to changes in the criteria for identification of bacteria as a result of development of modern diagnostic techniques. Multiple schemes for the identification of species of VGS have been proposed in literature. Colman and Williams (1972) classified VGS into five different species however later Facklam (1977) categorized VGS in 10 species. Lately, Ruoff (2002) divided VGS into 12 species characterized in 5 different groups namely; mutans group, salivarius group, bovis group, anginosus group and mitis group. Recently Facklam (2002) revised the classification and identification scheme of VGS and categorized into five different groups i.e., mutans group, anginosus group, mitis group, sanguinis group and salivarius group. Other than the five groups, *Streptococcus acidominimus* and *Streptococcus uberis* belong to genus *Streptococcus* but are considered other than VGS as miscellaneous streptococci.

MUTANS GROUP STREPTOCOCCI

Mutans group streptococci are the most important members of VGS. Mutans group streptococci includes *Streptococcus mutans* (Serotype c, e, f), *S.sobrinus* (Serotype d, g), *S. rattus* (Serotype b, formerly known as *S. ratti*), *S. cricetus* (Serotype a), *S. downei* (Serotype h), *S. macacae* (Serotype c), *S. ferus* and *S. hyovaginalis*. Among these species, *S.downei*, *S. macacae*, *S. ferus* and *S. hyovaginalis* have been isolated from animal while the remaining species have been recovered from humans (Oluwo *et al*., 2021; Facklam, 2002). Members of mutans group streptococci can be easily isolated from the human oral cavity (Ferrer *et al*., 2021; Tamura, 2008). *S. mutans* has ability to express two important characteristics which appear to be significant in their cariogenicity. The first important character is their ability to colonize tooth surfaces whereas the other involves their acidogenic potential (acidogenicity). The acidogenic potential of *S. mutans* leads to the process of demineralization of enamel. Another important character of *S. mutans* is their acid tolerance (aciduricity) as *S. mutans* have been reported as a strong acid tolerant oral flora. *S. mutans* has the ability to synthesize extracellular polysaccharides (glucans) from sucrose by glucosyltransferase (Cawson, 1991). The glucans help to provide attachment mechanism for adhesion to the surface of teeth. Besides synthesizing extracellular polysaccharide, *S. mutans* has ability to form functional amyloids. These amyloids contribute in the formation of dental biofilm (Oli *et al*., 2012). Furthermore, the virulence of *S. mutans* also depends on the glucan binding proteins (Gbps). Mainly four types of Gbps are expressed by *S. mutans* i.e. GbpA, GbpB, GbpC, and GbpD which are expressed on surface and are involved in the interactions with glucan synthesis due to their structure, immunological and functional qualities (Duque *et al*., 2011). In addition, the virulence of *S. mutans* also depends on two different enzymes i.e. NagB and GlmS, both enzymes are importance in the synthesis of cell wall of bacteria. In this connection, NagB decreases the virulence of *S. mutans* whereas GlmS increases the virulence property of *S. mutans* during carobohydrate metabolism (Kawada-Matsuo *et al*., 2012). Moreover, *S. mutans* has the ability to convert a variety of carbohydrates to different organic acids by glycolysis and accumulate on the surface of tooth containing dental plaque (Lam *et al*., 2022; Morimoto-Yamashita *et al*., 2011; Biswas and Biswas, 2012). *S. mutans* can tolerate the environmental stress such as pH and redox potential that are considered as virulence traits for the progression of dental caries. These environmental changes or altered conditions require a sophisticated and complex regulatory system as well as expression at genetic level to allow the bacteria to adapt altered environmental conditions for maintaining some important basic metabolic mechanisms for their survival (Bradshaw *et al*., 2002). However, the molecular mechanisms at gene expression level of the altered and adopted environment by *S. mutans* has not yet completely defined in the literature (Xue *et al*., 2010). In this regard, Svensater and group (2000) noted that bacterial cells can be protected against acid challenges at pH 3.5 by starvation stresses and adaptation to salts and acids. They further demonstrated an approach at proteomic level by mediating the stress responses after finding a significant level of altered expression of proteins under a variety of stress responses. Gong *et al*. (2009) also reported that approximately 14% of different genes could be differentially expressed in the genome during acidic pH. *S. mutans* has ability to give a quick response to or sense environmental acid fluctuations by using Two-component system (TCSs). The TCSs comprises a response of regulatory protein and sensor kinase that have an important role for the optimal growth of *S. mutans*(Tremblay *et al*., 2009). Besides, *S. mutans*has well-adapted acid defense mechanisms i.e. F1Fo-ATPases, malolactic fermentation (MLF) and agmatinedeiminase systems. The system of F1Fo-ATPases pumps the protons out from the cells and MLF and agmatinedeiminase systems help in the generation of ATP as an energy source and promote alkalization in pH of cytoplasm (Song *et al*., 2012). In addition, *S. mutans* lacks the heme-containing peroxidase or catalases which have the ability to degrade hydrogen peroxide (H_2O_2). Mainly *S. mutans* depends on superoxide dismutase, glutathione reductase and NADH oxidase activities for their protection from the reactive oxygen microbial species (Baldeck and Marquis, 2008). A phenotypic and transcriptomic study proved that oxygen is responsible for reducing biofilm formation and to cause alteration in the activity of sugar transportation and rate of glycolysis (Poorani *et al*., 2022; Xue *et al*., 2010).

Beside cariogenic role of mutans group streptococci particularly *S. mutans* and *S. sobrinus*, these species are also involved in other important diseases e.g. endocarditis (Brooks *et al*., 2002). Most of members of mutans group streptococci have also been reported as an important etiological agent of dental caries in human. An epidemiological study by Wu (2009) reported that *S. mutans* is associated with dental caries in humans in 90% cases followed by *S. sobrinus*.

ANGINOSUS GROUP STREPTOCOCCI

Another important group of VGS is the anginosus group strepotocci (previously reported as *S. milleri group*). This group consists of *S. anginosus*, *S. intermedius* and *S. constellatus* (Facklam, 2002). They are commensal inhabitants of the oral cavity, vaginal, genital and gastrointestinal tracts and are commonly associated with purulent infections in humans. The unique character of anginosus group streptococci is their involvement in abscesses in significantly higher percentages than other VGS (Pecharki *et al*., 2005). Besides, anginosus group streptococci are involved in endocarditis, purulent abdominal, catheter-related and neutropenia-related bloodstream infections, hepatobiliary, dental and brain infections (Whiley *et al*., 1992; Murray *et al*., 2007).

MITIS GROUP STREPTOCOCCI

Mitis group streptococci comprises *S. mitis*, *S. oralis* (previously famous as *S. sanguis* II), *S. crista* (previously known as *S. cristatus*) *S. infantis, S. peroris* and *S. orisratti*. All species have been isolated from human except *S. orisratti* (Facklam, 2002). Mitis group streptococci are prominent oropharyngeal microflora of human and responsible for causing bacterial endocarditis (Do *et al*., 2011), bacteremia (Achour *et al*., 2004), nosocomial blood stream infections (Lyytikainen *et al*., 2004), pneumonia (Smith *et al*., 2004), suppurative oral and maxillofacial infections, toxic shock syndrome (Tunkel *et al*., 2002), encephalopathy and adult respiratory distress syndrome (Alcaide *et al*., 1996). Among the members of mitis group, *S. oralis* is a virulent species and has the ability to synthesize sialidase. Sialidase (exo-glycosidase) is an enzyme that helps in multiplication and divisions of *S. oralis* during disease process (Byers *et al*., 2000).

SANGUINUS GROUP STREPTOCOCCI

The fourth important group of VGS is sanguinis group streptococci, which includes *S. sanguinis*(previously famous as *S. sanguis*), *S. parasanguinis* (formerly known as *S. parasanguis*) and *S. gordonii*. Most of the members of this group are recognized not only for their historical involvement in endocarditis but also due to their putative antagonistic characteristics in dental caries and periodontal disorders (Caufield *et al*., 2000). These species colonize tooth surfaces by formation of dental plaque and can be recovered from buccal mucosa (Frandsen *et al*., 1991; Volk *et al*., 1991).

SALIVARIUS GROUP STREPTOCOCCI

Salivarius group streptococci, the fifth group of VGS consists of *S. salivarius*, *S. infantarius*, *S. vestibularis*, *S. alactolyticus*, *S. hyointeninalis* and *S. thermophilus*. Among salivarius group, *S. salivarius*, *S. infantarius* and *S. vestibularis* have been reported to be isolated from human whereas *S. alactolyticus*, *S. hyointeninalis* and *S. thermophilus* have been obtained from animals. Salivarius group streptococci are normal inhabitant of tongue, mucosal surfaces and can be obtained from human saliva. They are primary colonizers in oral cavity of human after birth. *Streptococcus salivarius* is rarely responsible to cause any infection in humans except dental caries. It has unique ability to synthesize an extracellular polysaccharide and produce urease (Oluwo *et al*., 2021; Tamura *et al*., 2009).

MISCELLANEOUS SPECIES OF STREPTOCOCCI

STREPTOCOCCUS ACIDOMINIMUS

Streptococcus acidominimus belongs to genus *Streptococcus* but is considered other than VGS (Facklam, 2002). They are normal inhabitant of vaginal tract of bovine and calves' skin and present in raw milk. First report of isolation of *S. acidominimus* was from cows in 1922 (as cited in Ayers and Mudge, 1922). Formerly, it was rarely associated with human infections (Akaike *et al*., 1988) however recent literature have documented its isolation from abscess, wound, and genital tract of female individuals and characterized as a prominent member of VGS (Dalal and Urban, 2008). *S. acidominimus* is associated with endocarditis (Brachlow *et al*., 2003), pneumonia (Baker and Carlson, 2008), meningitis, pericarditis, gradenigo syndrome (acute petrositis) (Finkelstein *et al*., 2003), brain abscess, upper genital tract infections (Rabe *et al*., 1988) and massive ascites (Zhang and Qian, 2004). Especially in human, *S. acidominimus* is commonly involved in invasive infections (Baker and Carlson, 2008).

STREPTOCOCCUS UBERIS

S. uberis belongs to *Streptococcaceae* family that encompasses bacteria capable of pathogenic as well as commensal behavior. This group behaves as commensal in various tissues and has been recovered from the gut, skin, tonsils and genital tract of cattles. *S. uberis* do not have ability to produce extracellular polysaccharides. *S. uberis* is commonly responsible to cause bovine mastitis worldwide. They are also involved with pyrexial infections in animals (Khan *et al*., 2003). In humans, *S. uberis* is rarely responsible to cause infection but more recently, Gulen *et al*. (2012) reported few infections in humans isolated from urine samples of immunocompetent patients. Moreover, few other studies have discussed various human infections caused by *S. uberis* i.e. genital infections, pneumonia and joint infections (Kessel and Wittenberg, 2008; Khan *et al*., 2003).

DENTAL CARIES

Dental caries is a common prevalent disease and affects the quality of life. Despite the improvements in dental practices, dental caries still remains the most important disease globally. The prevalence of dental caries is increasing day by day in the developing and underdeveloped countries throughout the world.

Dental caries is predominant cause of tooth loss (especially crown of teeth) in children and young adults. It is the disintegration of tooth beginning at the surface and progressing inwards. Dental caries is an uncontrolled infection and will continuously progress if left untreated. Moreover, it is expensive to treat and not affordable by general public (Poorani *et al*., 2022; Bowen, 2016; Prashant *et al*., 2007; Sheiham, 2001; Balakrishnan *et al*., 2000).

Dental caries is a multifactorial disease categorized in three different factors i.e. risk factor, risk indicator and risk inhibitor. It is hard to determine one major factor associated with dental caries because they are all interlinked with each other. Risk factors are defined as any character that plays an important role in the progression of dental caries. Teeth, cariogenic bacteria, fermentable carbohydrate (sucrose) and exposure time are common risk factors. While, risk indicator is referred to co-existence of any character with high probability of development of dental caries or may show a measurable effect on health. In this respect, sociodemographic characteristics (i.e. age, sex, marital status and socioeconomic status) and other oral health related risk factors (dietary habits, chewing habits, use of betel quid products, use of tobacco, use of tea and oral hygiene practices) are included. Besides, risk inhibitor is designated as any character related with significant outcome of results of dental caries. Risk inhibitors include use of fluoride and buffering capacity and flow rate of saliva. These risk factors are also famous as caries promoting factors (risk factor and risk indicator) and caries inhibiting factors (risk inhibitor) (Lam *et al*., 2022; Paulander, 2004; Burt, 2005).

DENTAL PLAQUE

Dental plaque (microbial plaque or dental biofilm) is a unique type of complex biofilm that forms on the surfaces within the oral cavity. It consists of an organized and multi-diverse bacterial community which is enclosed within a matrix of variety of extracellular materials and attached to the dental surfaces. Dental plaque appears as a pale yellow or white slime layer which is commonly present between the teeth as well as along the cervical margins. The formation of dental plaque is natural phenomena that cannot be prevented. Removal of dental plaque is essential step for dental caries. As dental plaque becomes acidic, it helps to initiate the process of demineralization of tooth enamel or harden into dental calculus (Poorani *et al*., 2022). Calculus is commonly known as tartar. There are six steps in the formation of dental plaque such as association (pellicle forms on tooth and help bacteria to provide surface for attachment), adhesion (within short interval, bacterial flora firmly bind to the dental pellicle), proliferation (microbial flora spreads throughout the oral cavity and begin to multiply), microcolonies (VGS secrete slime layer as microcolonies are formed), biofilm formation (different complex bacterial groups are formed with metabolic advantages by microcolonies) and growth or maturation (the dental biofilm forms a primitive or unsophisticated circulatory system) (Lemos *et al*., 2019; Kreth *et al*., 2008; Marsh, 2003).

ROLE OF DENTAL PLAQUE IN DENTAL CARIES

Ecology of dental plaque might be modulated and influenced by several factors, like nutrient availability, population density, evasion of host immune components and diffusion of metabolites. The microbiological and biochemical composition of dental plaque can be changed in the presence of high intake of carbohydrates/ sugars, which transforms the healthy state of dental biofilm towards cariogenic dental biofilm as well as leads to increase growth proportion of pathogenic bacteria as compared to normal flora resulting in formation and development of cavities or dental caries (Sekundo *et al*., 2022). In the dental biofilm, a variety of microorganisms can be exposed to high amount of sugars/carbohydrates for short intervals and few of them are capable by utilizing carbohydrates to produce acid, store energy and synthesize soluble and insoluble extracellular polysaccharides. In short time, after exposure to carbohydrates, dental plaque undergoes in long periods of sugar starvation. These physiological changes of microbial growth are famous as famine or feast episodes which may be responsible to cause strategies related to microbiological selection that help to increase the number of acid tolerant bacterial species (*S. mutans*) in the dental biofilm (Ccahuana-Vasquez and Cury, 2010).

ROLE OF STREPTOCOCCI IN DENTAL CARIES

The oral VGS show important characteristics which appear to be significant in their ability to produce dental caries i.e. cariogenecity and acidogenic potential. The most important characteristic of VGS is to adhere to mucosa and tooth surface whereas second characteric of these bacteria the strong acidogenic potential which leads to demineralization of tooth enamel surface. The acidogenic bacteria utilize fermentable carbohydrate and form two substances i.e. organic acid and glucan (extracellular polysaccharide) (Lam *et al*., 2022; Ito *et al*., 2011; Arthur *et al*., 2011; Cawson, 1991).

The organic acid formed by VGS from fermentable carbohydrates diffuses into the tooth enamel and dentine and partially or completely dissolves the mineral form of crystals down inside the tooth. The tooth enamel and dentine are made up of millions of tiny crystals. The mineral involved is known as carbonated hydroxyapatite. This is calcium phosphate with numerous impurity inclusion, the most important of which is the bicarbonate ion which makes the mineral more acid soluble than pure hydroxyapatite. If the dissolving of the teeth mineral is not reversed, the primary subsurface lesions become a cavity. Generally dental caries occurs in permanent and deciduous teeth (regardless of the age of the subjects). A carious lesion at the very early stage occurs as a 'white spot', which progresses to frank open cavities when left untreated (Meric *et al*., 2020; Featherstone, 2003).

Besides organic acid, VGS also produce glucan (Extracellular polysaccharide). Glucan is a sticky or shiny gel, relatively inert and enhances plaque formation. It combines with components of saliva to form insoluble substance which adheres tenaciously to the surface of teeth and act as barrier against the diffusion of salivary buffer which is important in neutralization of acid produced by VGS. Glucan also helps cariogenic bacteria in the attachment to the surface of teeth. The evidence for the contribution of glucan in initiation of dental diseases especially dental caries has been proved by the fact that those strains of *Streptococcus mutans* which have lost the ability to form glucan become non-cariogenic (Cawson, 1991). Glucan also promote bacterial adherence to heart valve and subsequently vegetation formation. The vegetation formation is the rapid growth and persistence of bacteria embedded in platelet-fibrin thrombus, which progresses to subacute bacterial endocarditis (Brooks *et al*., 2002; Vriesema *et al*., 2000). The subacute bacterial endocarditis often involves abnormal valves (congenital deformities and rheumatic or atherosclerotic lesions). Although any organism reaching the blood stream may establish itself on thrombotic lesions that develop on endothelium injured as a result of circulator stresses, subacute endocarditis is most frequently due to members of VGS that have accidentally reached the blood, for example, after dental extraction at least 30% of patients have viridans streptococcal bacteremia, which is the cause of subacute bacterial endocarditis (Brooks *et al*., 2002).

GLUCAN PRODUCTION BY VGS

Glucan (extracellular polysaccharides) producing ability of VGS has considerable importance in the cariogenicity as well as in the formation of dental plaque (Ito *et al*., 2012). First reported discovery of EPS (extracellular polysaccharides) was done by Louis Pasteur in 1861. Later, it was designated as Glucan (dextran) after the determination of its empirical formula by Scheubler in 1874. Different microbial genera have potential to synthesize glucans such as *Leuconostoc*, *Streptococcus* and *Acetobacter* species (Qader *et al*., 2005). First reported research related to microbial production of glucan by *Leuconostoc* species was conducted by Hucker and Pederson in 1930. Another study (Hehre and Hamilton, 1949) reported glucan production by *Leuconostoc* species. Moreover, glucan production from dietary sucrose by *Streptococci* and *Acetobacter* species were reported in 1941 and 1954 by the teams of Niven and Jeanes, respectively.

Glucans are D-glucose polymers (polyglucosan) linked by glucosidic bonds. The production of glucans is catalyzed by glucosyltransferase enzymes (GTFs) using dietary sucrose. Generally, glucans are of two types i.e. soluble and insoluble. Glucans have difference in their physical and chemical characteristics such as molecular weight, solubility in water, intrinsic viscosity, specific rotation, nature of solid products and difference in the proportion of glucosidic bonds (glucopyranosidal units). According to molecular weight, soluble glucans are less in weight (20-50,000 Daltons) as compared to insoluble glucans (10^6-10^7 Daltons). Mainly, soluble glucans consist of α- 1, 6 glucosidic bond linkages whereas insoluble glucans comprise high numbers of α- 1, 3 glucosidic bonds linkages. Compared to soluble glucans, the insoluble glucans are recognized as virulence factor for the cariogenicity of various species of VGS and development of dental caries. The insoluble glucans play role as a barrier against the diffusion of salivary buffer which neutralize the acids formed in dental plaque by the activity of cariogenic species of VGS. Glucans are considered as an important virulence factors for cariogenicity of *S. mutans*. They can promote adherence of bacteria to the tooth surfaces. In addition, glucans consequently increase the demineralization of tooth enamel and have a major role in the porosity and structural integrity of biofilm. Glucans have been reported as an antigen and haptene (Sekundo *et al*., 2022; Ccahuana-Vasquez and Cury, 2010).

ROLE OF GLUCOSYLTRANSFERASES ENZYMES

The glucosyltransferases are sucrose metabolizing enzymes that play an essential role in the formation of dental plaque as well as in the sucrose dependent cellular adhesion. There are three different types of glucosyltransferase enzymes, GtfB, GtfC and GtfD. Among types of glucosyltransferases, GtfB and GtfC are responsible to synthesize insoluble glucans, both soluble and insoluble glucans whereas and only GtfD produces soluble glucans. GtfB and GtfC are of great concern with respect to the virulence of cariogenic organisms and pathogenesis of dental caries. Glucosyltransferases are active in nature and found on the surface of cariogenic microorganisms. They are also present in the saliva and salivary pellicle which formed on the surface of tooth in human oral cavity (Shemesh *et al*., 2006; Koo *et al*., 2003).

PREVENTATION & MANAGEMENT OF DENTAL CARIES

In past, the common approach for the treatment of dental caries was complete removal of diseased part of tissues and replacing them with an active restoration. But there was no attempt for curing of disease from this type of management and often individuals required more fillings after few months due to recurrent dental caries. In management of dental caries, the newer and latest philosophy highlights the worth of proper diagnosis, active prevention and techniques related to minimal cavity preparation (Kumar *et al*., 2018). The end point of these methodologies would require less instead of more restorative work by any patient (Jeevarathan *et al*., 2007). Since many years, different populations around the world are following various approaches for prevention of dental caries i.e., to stop sucrose consumption of carbohydrate between meal or to use non-cariogenic sweeteners such as sorbitol, xylitol and lycasin, to make the structure of tooth less soluble to acid attack by using fluoride products, to use sealants for protection of different areas of susceptible tooth such as pits and fissures, to minimize the acid production that would reduce or eliminate cariogenic bacteria in the presence of dietary sucrose (Sekundo *et al*., 2022; Kim and Lee, 2020). The acid production would be reduced by using antibiotics, possible immunization and oral hygiene aids. Although, the rate of prevalence of dental caries decreases using different preventive measures but still dental caries is the most prevalent disease all around the world. So there is a need for more curative and preventive dental care. Moreover, specific planning and oral awareness educational programs are implicated in order to prevent and control dental and oral diseases. These oral health based knowledge developing strategies should be deployed at home, community and school areas (Meric*et al*., 2020; Ferro *et al*., 2009; Jeevarathan*et al*., 2007; Al-Malik and Rehbini, 2006).

DIET AND DENTAL CARIES

A proper diet can also play an essential role in removing dental plaque because structure and condition of teeth are dependent on dietary habit of mankind (Kutsh and Young, 2011). Diet containing sweets and refined food items which are made by white flour and sugars are very harmful as compared to raw vegetables and whole food products. The whole food products are known as detergent food products such as millet, sesame seeds and onions. These products are very beneficial for oral health because of their ability to remove dental plaque from teeth and firm the gums and teeth (Kumar *et al*., 2018; Thanyasrisung *et al*., 2009).

Dental caries is a diet dependent disease. The use of dietary sucrose is considered to be a key factor in promoting the dental caries (Makinen, 2011). Sucrose increases the risk of dental caries than other types of sugars. This concept has been reviewed by researchers that dental caries can be restricted by using and substituting non-caries promoting constituents in routine diet. Few examples of these products are hydrogenated starch, sorbitol, xylitol and fructose. Except xylitol, all sugars markedly increase the prevalence of dental caries (Schneider *et al*., 2012; Assev *et al*., 2002). In this respect, the concentration and amount of sugar in diet, physical form of sugar, oral retentiveness (a length of time when teeth are exposed to decrease level of pH in dental plaque), length of interval between eating, sequence of food consumption, drinking ratio and frequency of eating meals and snacks can play a major role in the process of dental caries. Generally if sugar is taken only at meal time then the pH of dental plaque critically drops for 3 hours per day while if sugar is used between meals than pH becomes lower than its critical level. The high consumption of vegetables, whole fruits, starchy staple foods, milk and water to replace sugary food and drink products promotes remineralization process and decrease the risk of dental caries (Kumar *et al*., 2018; Malik *et al*., 2013; Harties and Leach, 1975).

Moreover, oral VGS have the ability to ferment intrinsic and extrinsic sugars to acids which dissolve the tooth enamel. Both intrinsic and extrinsic based sugar products promote dental caries. Intrinsic is termed as natural food products that are present within the cellular body of food such as fruits and vegetables whereas extrinsic sugars are those which present are free in food or artificial addition of sugar. The extrinsic sugar products are further grouped into milk (lactose) (MES) and non-milk (honey and added sugars) extrinsic sugars (NEMS). Malik *et al*. (2009) reported that high consumption of NMES may promote risk of dental caries. They can increase the prevalence of obesity as well as weight gain. The artificial sweeteners can not be metabolized and absorbed by cariogenic bacteria for production of acid. There are two types of sugar sweeteners such as nutritive sweeteners (sugar alcohols, sorbitol, lycasin, and xylitol) and non-nutritive sweeteners (aspartame, sorbitol). In this connection, sugar sweetened beverages (SSB) are also harmful for oral health. SSB are those drinks which have artificially added sugar such as non-diet sodas or soft drinks, sports drinks, flavored juice drinks, coffee drinks, sweetened tea, electrolyte replacement and energy drinks. The calories present in SSB do not have nutritional value and can contribute to weight gain. It leads to dental caries, obesity, diabetes and heart disease. Literature has been discussed that these SSB products are responsible for increasing the risk of dental caries about 80-100% among children belonging to 1-5years age (Marshall *et al*., 2003). An Australian based research carried out by Cochrane *et al*. (2012) found the protective effect of sugar free chewing gums that have the ability to promote the process of remineralization and decrease the risk of dental caries. Many other scientists (Chestnutt *et al*., 2003; Moynihan and Petersen, 2004; Sohn*et al*., 2006; Delpier *et al*., 2013) also supported the drawback of use of high level of SSB products for dental caries.

SUCROSE AND DENTAL CARIES

Dental caries is related to consumption of dietary carbohydrates that are the most important risk factor. Dietary carbohydrate is used as substrate in the process of dental caries. In this respect, sucrose and starches are the major components present in different forms in diet. Comparatively, starch is a high molecular weight carbohydrate than sucrose and not easily available for bacteria because bacteria cannot easily break to smaller units. It is an essential step for formation of dental plaque. Generally sucrose is easily available for bacterial flora. It is famous as the key substance for dental caries process. It is evident from different literatures that high consumption of dietary sucrose can promote and increase dental caries but the prevalence of dental caries may also depend on manners for administration of consumed dietary sucrose. VGS have the ability to utilize dietary sucrose as a nutrient source. Risk of dental caries can be minimized by changing dietary habits of human (Meric *et al*., 2020; Jeevarathan *et al*., 2007).

ORAL HYGIENE AND DENTAL CARIES

Generally, human health depends on oral health as it affects the complete well-being of human. Good hygienic condition of oral cavity is a key factor for preventing dental caries and improving the oral and dental health. Dental caries is one of the most common and expensive disease related to mankind. Different types of remedies are used for the removal of dental plaque and dental caries. Proper cleaning of teeth is essential for maintaining oral health. The mechanical removal of plaque by tooth brushing and flossing can almost completely prevent caries and periodontal disease (Marcotte and Eavoie, 1998). The process of tooth brushing is one of the best, timeless and cost effective tools for cleaning of teeth. Generally, frequent brushing i.e. minimum twice a day is essential and recommended for cleaning teeth and gums at the early age. Moreover, frequently brushing with fluoride containing toothpastes can have a great impact on teeth and oral health. Furthermore, regular checkup is also important (Meric *et al*., 2020; Assev *et al*., 2002).

FLUORIDE AND DENTAL CARIES

One of the famous and most cost effective ways of reducing dental decay is water fluoridation. At level of 1ppm, fluoride is added in the water can reduce up to 60% prevalence of dental caries during dental development. Alternatively to water fluoridation, addition of fluoride in table salt or milk might also reduce dental caries but it is not effective than fluoridation of water. Fluoride is absorbed more quickly from water than from milk. For children, addition of fluorides in milk (milk fluoridation) increases the pH of dental plaque, decreases the ratio of cariogenic organism and increases the proportion of non-cariogenic bacteria. Fluoridated tooth picks have also been reported to inhibit demineralization of tooth enamel. Besides, during dental development, fluoride is being effectively used in tablets, mouth rinses and oral drops reduce dental caries but doses of tablets must be controlled carefully due to risk of dental fluorosis (mottling). Dental fluorosis is a specific defect of tooth enamel because of consumption of excess amount (over 2ppm) of fluoride during dental development. Moreover, for direct topical application, acidulated phosphate or sodium fluorides (fluoride varnish) can also be used to the teeth among individuals with high risk of dental caries. The actual mechanism of protection by use of fluoride is doubtful or questionable but fluoride may incorporate into the tooth enamel as fluoraptite and make it more resistant toward acid attack and enhance the process of remineralization. It may interfere with metabolism of bacteria in the dental plaque. Furthermore, fluoride containing dentifrices such as fluoride toothpastes are commonly used for reducing dental plaque formation (tartar) and prevent from dental caries and improve gum health (Jeevarathan *et al*., 2007).

ANTIBIOTIC AND THEIR RESISTANCE

Generally, antibiotics are being used in dental procedures to treat prophylaxis, local infections and odontogenic and nonodontogenic infections. The most common antibiotics used are Penicillin, tetracyclines, amoxicillin, clindamycin, metronidazole, ciprofloxacin, and azithromycin (Chun and Lee, 2015). They are active against various microorangisms, and worked particularly well to control against dental pain because they block the enzyme that causes gums to become swollen. Antibiotics are also being used in different ways for various purposes such as animal feed additives, promoting plant growth and self medication. Besides, high potencies of antibiotics are mostly used for treatment of minor infections and more than one antibiotic is prescribed by the doctors for a single infection (Kim and Lee, 2020; Chun and Lee, 2015).

Evident from various citations that antibiotic plays an important role in the treatment and control of various infections and diseases. But we can not ignore an importance of antibiotic resistance that is a major health problem worldwide. All types of antibiotic classes have extra-chromosomal genes that may play a vital role in antibiotic resistance. These extra-chromosomal genes contain an extra, transferable, large chromosomal DNA element. Besides, integrons and tansposons are also present on extrachromosomal plasmid. Integrons and transposons are mobile DNA elements and play a vital role in the acquisition of antibiotic resistance. They are helpful in rapid dissemination related resistance gene transformation of genetic determinants among various bacteria (McDermotte *et al*., 2002). Thus increasing rate of development of resistant organisms to different antibiotics promotes an urgent requirement for newer synthetic or modified antibiotics (Tollfson and Miller, 2000). The indiscriminate use of synthetic antibiotics promotes the increase in the number of multidrug resistant bacteria (MDR). Thus treatment and prevention of infection related to these bacteria are ultimately too difficult (Kim and Lee, 2020; McDermott *et al*., 2002).

NATURAL SUBSTANCES AND DENTAL CARIES

Due to increase in multi-drug resistance and side effect of used antibiotic, there is a need of alternative, curative and preventive measures which should also be safe, economical and beneficial for mankind. Uncountable parts of plants, herbs and spices have been used for many years in prevention of dental caries (Ferrazzano *et al*., 2011). They are easily available and in domestic setting can be used in self medication. Polyphenoloxidase (PPO) is an enzyme found usually in non-citrus fruits and many vegetables has been reported effective against colonization of oral bacteria, for instance, *S. sorbinus*. Natural substances like chewing sticks i.e. miswak is more often used as a mechanical and chemical cleansing of tooth tissues compared to a toothbrush. Chemical constituents and minerals such as sodium, chloride, potassium and bicarbonate fight plaque and gum infections, as well as give strength the enamel of teeth and prevent from any existing teeth decay from getting worse. Moreover, hinokitiol, cinnamon bark oil, papua-mace extracts and clove bud oil in spice extracts (Saeki *et al*., 1989) and some other spices like thyme, sage, tansy, oregano and peppermint were also found effective against oral flora (Woodward, 1999). In addition, some of the other oils showed potent activity against anaerobic oral bacteria like Australian tea tree oil, peppermint oil and sage oil. Thymol, the component of essential oils of sage and oregano was also found effective against oral bacteria.

CONCLUSION

Indeed, importance of oral flora i.e. viridans group streptococci cannot be ignored with respect to dental caries because complete general health is not possible to achieve without curing oral health, so proper preventive measures should be used that are mostly mentioned in present study. Obviously, treatment of dental caries is very costly, normally general public cannot afford the expense but with simple preventive measures dental caries can be easily controlled such as brushing, flossing (tooth cleaning daily with toothpaste, toothpowder, chewing sticks and mouthwashes) and dietary habits (proper use of sugar containing items or a diet low in sugar).Within the limitations of present study, it could be served as a baseline for awareness about prevalence and trend of dental caries at regional level especially in Pakistan.

REFERENCES

Achour, W., O. Guenni, B.Malbruny, A.Canu, R. Leclerco and A. B.Hassen (2004). Phenotypic and molecular characterization of macrolide and streptogramin resistance in *S. mitis* from neutropenic patients.*J AntimicrobChemother*., 54: 117-121..

Akaike, T., M. Suga, M., Ando, Y. Ando, S. Araki and R. Fujise (1988).*Streptococcusacidominimus* infections in a human. *Jpn J Med*., 27: 317-320.

Alcaide, F., J. Linares and R. Pallares (1995).*In vitro* activities of 22- Lactam antibiotics against penicillin-resistant and penicillin-susceptible viridans group streptococci isolated from blood. *Journal of Antimicrobial Chemotherapy*, 39: 2243-7.

Al-Malik, M.I, and Y. A.Rehbini (2006). Prevalence of dental caries, severity, and pattern in age 6 to 7-year-old children in a selected community in Saudi Arabia. *J Contemp Dent Pract*., 7(2): 046-054.

Arthur, R. A.,A. A. Cury, R. O. Graner, P. L.Rosalen, G. C. Vale, A. F. Paes Leme, J. A. Cury and C. P. Tabchoury (2011). Genotypic and phenotypic analysis of *S. mutans* isolated from dental biofilms formed *in vivo* under high cariogenic conditions. *Brazilian Dental Journal*, 22(4): 267-74.

Assev, S., S. Stig and A. A. Scheie (2002).Cariogenic traits in xylitol-resistant and xylitol-sensitive mutans streptococci.*Oral Microbiol Immunol*., 17: 95-99.

Ayers, J. S. H. and C. S. Mudge (1922).The streptococci of the bovine udder. IV. Studies of the streptococci. *Journal of Infectious Diseases*, 31: 40-50.

Baker, L. and R. Carlson (2008).*Streptococcus acidominimus* isolated from a multiloculated empyema in a critically ill adult man with pneumonia: case report and review of literature.*Heart and Lung*, 37(4): 308-10.

Balakrishnan, M., R. S. Simmonds and J. R. Tagg (2000). Dental caries is a preventable infectious disease. *Australin Dental Journal*, 45(4): 235-245.

Baron, E. J., L.R. Peterson and S.M. Finegold (1994).Bailey & Scott's, Diagnostic Microbiology, 9^{th} Edition. The C.V Mosby Company, pp. 333-351.

Belda-Ferre, P., L. D. Alcaraz, R. Cabrera-Rubio, H. Romero, A. Simon-Soro, M. Pignatelli and A. Mira (2012). The oral metagenome in health and disease.*ISME J*., 6(1): 46-56.

Baldeck, J. D. and R. E. Marquis(2008). Targets for hydrogen-peroxide-induced damage to suspension and biofilm cells of *S. mutans*. *Can J Microbiol*., 84: 868-875.

Bjorklund, A. B., B.A. Cook, B. R. Hendel-Peterson, P. F. Walker, W. M. Stauffer and D. R.Boulware (2011). Impact of global health residency training on medical knowledge of immigrant health.*Am J Trop Med Hyg*., 85(3): 405-8.

Blandino, G., I. Milazzo, D. Fazio, S. Puglisi, M. Pisano, A. Speciale and S. Pappalardo (2007). Antimicrobial susceptibility and beta-lactamase production of anaerobic and aerobic bacteria isolated from pus specimens from orofacial infections. *Journal of Chemotherapy*, 19(5): 495-9.

Bowen, H.W. (2016). Dental caries- not just holes in teeth: A perspective. *Mol Oral Microbiol*., 31(3):228-33.

Brachlow, A., S. Awadallah and A. Chatterjee (2003). Endocarditis due to *Streptococcus acidominimus*. *Pediatr Cardiol*., 24: 161-163.

Bradshaw, D. J., P. D. Marsh, R. J. Hodgson and J. M. Visser (2002). Effects of glucose and fluoride on competition and metabolism within *in vitro* dental bacterial communities and biofilms. *Caries Research*, 36: 81-86.

Brook, I. (1994). Micraerophilic streptococcal infection in children. *J. Infect*., 28(3): 241-9.

Brooks, G. F., J. L. Butel and L. N. Orston (2002). *Jawetz, Melnick and Adelberg's, Medical Microbiology*, 22nd Edition, 177, 178, 207, 208.

Burt, B. A. (2005). Concepts of risk in dental public health.*Community Dental and Oral Epidemiology*, 33: 240-7.

Byers, H. L., E. Tarelli, K. A. Homer and D. Beighton (2000). Isolation and characterization of sialidase from a strain of *Streptococcus oralis. Journal of Medical Microbiology*, 49: 235-244.

Caufield, P. W., A. P. Dasanayake, Y. Li, Y. Pan, J. Hsu and J. M. Hardin (2000). Natural history of *Streptococcus sanguinis* in the oral cavity of infants: Evidence for a discrete window of infectivity. *Inf. Immun*., 68(7): 4018-4023.

Cawson, R. A. (1991). *Essential of dental surgery and pathology*.5th edition.Longman Singapore publishers (Pvt) Ltd., pp. 31-174.

Ccahuana-Vasquez, R. A. and J. A. Cury (2010).*S. mutans* biofilm model to evaluate antimicrobial substances and enamel demineralization. *Braz. Oral Res*., 24(2): 135-41.

Chang, W. N., S. D. Chen, C. C. Lui, C. R. Huang and C. H. Lu (2003).Septic cavernous sinus thrombosis due to *Streptococcus constellatus* infection. *J.Formos Med. Assoc*., 102(10): 733-736.

Chattoraj, S. S., S. Ganesan, A. M. Jones, J. M.Helm, A. T.Comstock, R. Bright-Thomas, J. J. LiPuma, M. B. Hershenson and U. S. Sajjan (2011). Rhinovirus infection liberates planktonic bacteria from biofilm and increases chemokine responses in cystic fibrosis airway epithelial cells. *Thorax*, 66(4): 333-9.

Chestnutt, I. G., C. Murdoch and K. E. Robson (2003). Parents and carers' choice of drinks for infants and toddlers, in areas of social and economic disadvantage. *Community Dental Health*, 20(3): 139-45.

Chun, S., H, H. J. and N. Y. Lee (2015).Species-specific difference in antimicrobial susceptibility among viridans group streptococci. *Ann Lab Med*., 35(2):205-11.

Cochrane, N. J., P. Shen, S. J. Byrne, G. D. Walker, C. G. Adams, Y. Yuan, C. Reynolds, B. Hoffmann, S. G.Dashper and E. C. Reynolds (2012). Remineralisation by chewing sugar-free gums in a randomised controlled in situ trial including dietary intake and gauze to promote plaque formation. *Caries Research*, 46(2): 147-55.

Colman, G. and R. E. O. Williams (1972). *Taxonomy of some human viridans streotococci*, 281-299.In Wannamaker, L. W. and J. M. Matsen (ed.), *Streptococci and streptococcal diseases, recognition, understanding, and management*. Academic Press Inc., New York.

Dalal, A. and C. Urban (2008). Human Infections Due to *Streptococcus acidominimus*. *Infect. Dis.Clin.Pract*.,16: 283-284.

Delpier, T., S. Giordanaand B. M. Wedin(2013).Decreasing sugar-sweetened beverage consumption in the rural adolescent population. *J Pediatr Health Care*.,27(6): 470-8.

Do, T., S. C. Gilber, J. Klein, S. Warren, W. G. Wade and D. Beighton (2011). Clonal structure of *Streptococcussanguinis* strains isolated from endocarditis cases and the oral cavity. *Mol Oral Microbiol*., 26(5): 291-302.

Duque, C., R. N. Stipp, B. Wang, D. J. Smith, J. F.Hofling, H. K. Kuramitsu, M. J. Duncan and R. O. Mattos-Graner (2011). Downregulation of GbpB, a component of the VicRKregulon, affects biofilm formation and cell surface characteristics of *S. mutans*. *Infection and Immunity*, 79(2): 786-796.

Edson, R. S., D. R. Osmon and D. J.Berry (2002). Septic arthritis due to *Streptococcus sanguis*. *Mayo Clin. Proc*., 77(7): 709-10.

Elavarasu, S., P. Jayapalan and T. Murugan (2012). Bugs that debugs: Probiotics. *J Pharm Bioallied Sci*., 4(Suppl 2): S319-22.

Ergin, A. (2010). Classical and new approaches in laboratory diagnosis of viridans streptococci. *Mikrobiyol Bul*., 44(3): 495-503.

Fackalm, R. (2002). What happened to the streptococci: overview of taxonomic and nomenclature changes. *Clinical Microbiological Reviews*, 15: 613-30.

Facklam, R. R. (1977). Physiological differentiation of viridans streptococci. *J Clin.Microbiol*., 5(2): 184-201.

Fang, P. H., W. C. Lin, N. W. Tsai, W. N. Chang, C. R. Huang, H. W.Chang, T. L. Huang, H. C. Lin, Y. J. Lin, B. C. Cheng, B. Y. Su, C. T. Kung, H. C. Wang and C. H. Lu (2012). Bacterial brain abscess in patients with nasopharyngeal carcinoma following radiotherapy: microbiology, clinical features and therapeutic outcomes. *BMC Infect Dis*., 12: 204.

Featherstone, J. D. (2008). Dental caries: a dynamic disease process. *Australian Dental Journal*, 53: 286-291.

Ferrazzano, G. F., I. Amato, A.Ingenito, A. De Natale and A. Pollio (2011). Anti-cariogenic effects of polyphenols from plant stimulant beverages (cocoa, coffee, tea). *Fitoterapia*, 80: 255-262.

Ferrer, M.D., S. Perez, A. L. Lopez, J. L. Sanz, M.Melo, C.Liena and Mira (2021). Evaluation of Clinical, Biochemical and Microbiological Markers Related to Dental Caries.*Int J Environ Res Public Health*, 18(11):6049.

Ferro, C. R., D. C. Oliveira, F. P.Nunes and L. S. Pieqas (2009). Postoperative atrial fibrillation after cardiac surgery. *Arq Bras Cardiol*., 93(1): 59-63.

Finkelstein, Y., N. Marcus, R.Mosseri, Z. Bar-Sever and B. Z. Garty (2003). *Streptococcus acidominimus i*nfection in a child causing Gradenigo syndrome. *Int J Pediatr Otorhinolaryngol*., 67(7): 815-7.

Frandsen, E. V., V. Pedrazzoli and M. Kilian (1991). Ecology of viridans streptococci in the oral cavity and pharynx.*Oral MicrobiolImmunol*., 6(3): 129-133.

Gong, Y., X. L. Tian, T. Sutherland, G. Sisson, J. Mai, J. Ling and Y. H. Li (2009). Global transcriptional analysis of acid-inducible genes in *Streptococcus mutans*: multiple two-component systems involved in acid adaptation. *Microbiology*, 155: 3322-3332.

Gülen, D. K. A., M. Aydin and Y. Tanriverdi (2012). Urinary tract infections caused by *Streptococcus uberis*: a pathogen of bovine mastitis-report of seven cases. *Afr J Microbiol Res*., 7(30): 3908-12.

Hardie, J. M. (1992). Oral microbiology: Current concepts in the microbiology of dental caries and periodontal disease. *Brazilian Dent Journal*, 172(7): 271-278.

Harrell, T. and J. S. Hammes (2012). Meningitis admitted to a military hospital: a retrospective case series. *Mil Med*., 177(10): 1223-6.

Harties, R. L. and S. A. Leach (1975). Effect of diet on dental caries.*Br Med Bull.*, 31(2): 137-41.

Hehre, E. J. and D. M. Hamilton (1949).Bacterial conversion of dextrin into polysaccharide with the serological properties of dextran. *ProcSocExptBiol Med N Y*., 71: 336-339.

Hillman, N. H., J. J. Pillow, M. K.Ball, G. R. Polglase, S. G. Kallapur and A. H. Jobe (2009). Antenatal and postnatal corticosteroid and resuscitation induced lung injury in preterm sheep. *Respir Res*., 10: 124.

Huang, D. F., C. Y. Tsai, R. S. Liu, A. H. Yang and C. D. Chou (2000). Reiter’s syndrome caused by *Streptococcus viridans* in a patient with HLA-B27 antigen. *Clin Exp Rheumatol*., 18(3): 394-396.

Hucker, G. J. and C. S. Pederson (1930). Studies on the coccaceae XVI.Genus Leuconostoc.*N Y AgrExptSta Tech Bull*., 167: 3-8.

Ito, A., M. Hayashi, T.Hamasaki and S. Ebisu (2011). Risk assessment of dental caries by using Classification and Regression Trees. *Journal of Dentistry*, 39(6): 457-63.

Jeevarathan, J., A. Deepti, M. S. Muthu, V. RathnaPrabhu and G. S. Chamundeeswari (2007). Effect of fluoride varnish on *Streptococcus mutans* counts in plaque of caries-free children using Dentocult SM strip mutans test: a randomized controlled triple blind study. *J Indian SocPedodPrev Dent*., 25(4): 157-63.

Kawada-Matsuo, M., Y. Mazda, Y. Oogai, M. Kajiya, T. Kawai, S. Yamada, S. Miyawaki, T. Oho and H. Komatsuzawa (2012). GlmS and NagB regulate amino sugar metabolism in opposing directions and affect *Streptococcus mutans* virulence. *PLoSONE*, 7(3): e33382.

Kessel, S. and C. E. Wittenberg (2008). Joint infection in a young patient caused by *Streptococcus uberis*, a pathogen of bovine mastitis- a case report. *Z OrthopUnfall*., 146(4): 507-9.

Khan, I. U., A. A. Hassan, A.Abdulmawjood, C. Lammler, W. Wolter and M. Zschock (2003). Identification and epidemiological characterization of *Streptococcus uberis* isolated from bovine mastitis using conventional and molecular methods. *Journal of Vetenairy Sciences*, 4(3): 213-223.

Kim, Y.H. and S. Y. Lee (2020). Antibiotic Resistance of Viridans Group Streptococci Isolated from Dental Plaques. *Biocontrol Sci*., 25(3):173-178.

Koo, H., P. L. Rosalen, J. A. Cury, Y. K. Park and W. H. Bowen (2003). Effects of compounds found in propolis on *Streptococcus mutans* growth and on glucosyltransferase activity. *Antimicrobial Agents and Chemotherapy*, 46(5): 1302-1309.

Kreth, J., L. Zhu, J. Merritt, W. Shi and F. Qi (2008). Role of sucrose in the fitness of *Streptococcus mutans*. *Oral MicrobiolImmunol*., 23: 213-219.

Kumar, V.N., M. Krishnamurthy, S. Poorni , S. Patil and A. T. Raj (2018). Probiotics in Caries Prevention. *J Contemp Dent Pract*., 19(2):123-124.

Kuribayashi, M., Y. Kitasako, K.Matin, A. Sadr, K. Shida and J. Tagami (2012). Intraoral pH measurement of carious lesions with qPCR of cariogenic bacteria to differentiate caries activity. *Journal of Dentistry*, 40(3): 222-8.

Kutsch, V. K. and D. A. Young (2011).New directions in the etiology of dental caries disease. *J Calif Dent Assoc*., 39(10): 716-21.

Lam, P.P.Y., H. Chua, M. Ekambaram, E. C. M. Lo and C.K.Y. Yiu (2022). Does Early Childhood Caries Increase Caries Development among School Children and Adolescents? A Systematic Review and Meta-Analysis. *Int J Environ Res Public Health*., 19(20):13459.

Law, V., W. K. Seow and G. Townsend (2007). Factors influencing oral colonization of *mutans streptococci* in young children. *Aust Dent J*., 52(2): 93-100.

Lemos, J.A., S. R. Palmer, L.Zeng, Z. T. Wen, J. K. Kajfasz, I. A. Freires, J. Abranches and L. J. Brady (2019). The biology of *S. mutan*.*MicrbiolSpectr*., 7(1):101-128.

Liu, L., H. Tong and X. Dong (2012). Function of the pyruvate oxidase-lactate oxidase cascade in interspecies competition between *Streptococcus oligofermentans* and *Streptococcus mutans*. *Applied Environmental Microbiology*, 78(7): 2120-7.

Lyytikainen, O., M. Rautio, P. Carlson, V. J. Anttila, R.Vuento, H. Sarkkinen, A. Kostiala, M. L.Vaisanen, A.Kanervo and P. Ruutu (2004). Nosocomial bloodstream infections due to viridans streptococci in haematological and non-haematological patients: species distribution and antimicrobial resistance. *Journal of Antimicrobial Chemotherapy*, 53: 631-634.

Maeda, Y., C. E. Goldshmith, W. A. Coulter, C. Mason, J. S. Dooley, C. J. Lowery, B. C. Millar and J. E. Moore (2011). Comparison of minimum inhibitory concentration by broth microdilution testing versus standard disc diffusion testing in the detection penicillin, erythromycin and ciprofloxacin resistance in viridans group streptococci. *Journal of Medical Microbiology*, 60(Pt 12): 1782-6.

Makinen, K. (2011). Sugar alcohol sweeteners as alternatives to sugar with special consideration of xylitol. *Med PrinPract*., 20(4): 303-20.

Malik, A., A. Qureshi and M. S. Shaukat (2013). Prevention of dental decay: role of fluoride in dentifrices. *J Dow Uni Health Sci*., 7(2): 59-62.

Marcotte, H. and M. C. Lavoie (1998). Oral microbial ecology and the role of salivary immunoglobulin A. *Microbiol Mol Biol Rev.*, 62: 71-109.

Marsh, P. D. (2003). Are dental diseases examples of ecological catastrophes? *Microbiology*, 149: 279-294.

Marshall, T. A., S. M. Levy, B.Broffitt, J. J. Warren, J. M. Eichenberger-Gilmore, T. L. Burns and P. J. Stumbo (2003). Dental caries and beverage consumption in young children. *Pediatrics*, 112(3): e184-e191.

Martinez-Martinez, R. E., R. A. Dominguez-Perez, J. Sancho-Mata, C. Abud-Mendoza, J. L. Avala-Herrera and E. A. Popoca-Hernandez (2019). The frequency and severity of dental caries, and counts of cariogenic bacteria in rheumatoid arthritis patients. *Dent Med Probl*., 56(2):137-142.

McDermott, P. F., S. Zhao, D. D. Wagner, S. Simjee, R. D. Walker and D. G. White (2000). The food safety perspective of antibiotic resistance. *J AOAC Int*., 83(2): 245-54.

Meric, E., B. Bolgui, N. Duran and E. Ay (2020). Evaluation of oral streptococci in saliva of children with severe Early Childhood Caries and caries-free. *Eur J Paediatr Dent*., 21(1):13-17.

Morimoto-Yamashita, Y., M. Matsuo, H. Komatsuzawa, K. Kawahara, K. Kikuchi, M. Torii and M. Tokuda (2011). MK615: A new therapeutic approach for the treatment of oral disease. *Med Hypotheses*., 77(2): 258-60.

Moynihan, P. and P. E. Petersen (2004). Diet, nutrition and the prevention of dental diseases, *Public Health and Nutrition*, 7: 201-226.

Murray, P. R., E. J. Baron, J. H. Jorgensen, M. A. Pfaller and M. L. Landry (2007). *Manual of Clinical Microbiology*, 9th Edition, pp. 412-426.

Niven, C. F. J. R., K. L. Smiley and J. M. Sherman (1941). The production of large amount of a polysaccharide by *Streptococcus salivarius*. *Journal of Bacteriology*, 41: 479-484.

Oli, M. W., H. N. Otoo, P. J. Crowley, K. P. Heim, M. M. Nascimento, C. B. Ramsook, P. N. Lipke and L. J. Brady (2012). Functional amyloid formation by *Streptococcus mutans*. *Microbiology,* 158: 2903-2916.

Oluwo, A.O., F. O. Nwakorie, F. A. Oredugba and E. O. Sote (2021). Comparative Analysis of *Streptococcus mutans* and *Streptococcus sobrinus* from Dental Plaque Samples of Nigerian Pre-school Children with and Without Caries. *West Afr J Med*., 38(10):972-978.

Ono, T., S. Shiota, K. Hirota, K. Nemoto, T. Tsuchiya and Y. Miyake (2000). Susceptibilities of oral and nasal isolates of *Streptococcus mitis* and *Streptococcus oralis* to macrolides and PCR detection of resistance genes. *Antimicrobial Agents and Chemotherapy*, 44: 1078–80.

Pasteur, L. (1861). Surla Fermentation Visquese et al., Fermentation Butyrique. *Bull Soc Chin Paris*., 11: 30-31.

Paulander, J. (2004). *Oral epidemiological studies in a Swedish adult population*. Thesis. Göteborg: University of Gothenburg.

Pecharki, D., F. C. Petersen, S. Assev and A. A. Scheie (2005). Involvement of antigen I/II surface proteins in *Streptococcus mutans* and *Streptococcus intermedius* biofilm formation. *Oral Microbiology and Immunology*, 20(6): 366-71.

Poorni, S., M. S. Nivedhitha, M. R. Srinivasan and A. Balasubramaniam (2022). Estimating Genotypic Diversity of S*treptococcus mutans*Isolated From Caries-Active and Caries-Free Individuals Among Indian Population. *Cureus*., 14(2):e22436.

Prashant, G. M., G.N.Chandu, K. S. Murulikrishna and M. D. Shafiulla (2007). The effect of amgo and neem extract on four organisms causing dental caries: *S. mutans*, *S. salivarius*, *S. mitis*, and *S. sanguis*: An *in vitro* study. *Indian J Dent Res*., 18(4): 148-51.

Presterl, E., A. J. Grisold, S. Reichmann, A. M. Hirschl, A. Georgopoulos and W. Graninger (2005). Viridans streptococci in endocarditis and neutropenic sepsis: biofilm formation and effects of antibiotics. *Journal of Antimicrobial Chemotherapy*, 55: 45-50.

Qader, S., A., L. Iqbal, A. Aman, E. Shireen and A. Azhar (2005). Production of dextran by newly isolated strains of *Leuconostocmesenteroides* PCSIR-4 and PCSIR-9.*Turkish Journal of Biochemistry*, 31(1): 21-26.

Rabe, L. K., K. K. Wintercheid and S. L. Hillier (1988). Association of viridans streptococcus from pregnant women with bacterial vaginosis and upper genital tract infections. *Antimicrobial Agent Chemotherapy*, 43(7): 1595-1599.

Ruoff, K. L. (2002). Miscellaneous catalase negative, gram-positive cocci: Emerging opportunists. *Journal of Clinical Microbiology*, 40(4): 1129-1133.

Saeki, Y., Y. Ito, M. Shibata, Y. Sato, K. Okuda and I. Takazoe (1989). Antimicrobial action of natural substances on oral bacteria. *Bull Tokyo Dent Coll*., 30(3): 129-135.

Schneider, M., G.Kirfel, M. Berthold, M. Frentzen, F. Krause and A. Braun (2012).The impact of antimicrobial photodynamic therapy in an artificial biofilm model. *Lasers Med Sci*., 27(3): 615-20.

Sekundo, C., E. Langowski, D. Wolff, S. Boutin and C. Frese (2022). Maintaining oral health for a hundred years and more? - An analysis of microbial and salivary factors in a cohort of centenarians. *J Oral Microbiol*., 14(1):2059-891.

Sheiham, A. (2001). Dietary effects on dental diseases. *Public Health Nutr*.,4(2B): 569-91.

Shelburne, S. A., S. B. Greenberg, S. Aslamand D. J. Tweardy(2007).Successful ceftriaxone therapy of endocarditis due to penicillin non-susceptible viridans streptococci.*J Infect*., 54(2): e99-101.

Shemesh, M,, A. Tam, M. Feldman and D. Steinberg (2006). Differential expression profiles of *Streptococcus mutansftf*, *gtf* and *vic*R genes in the presence of dietary carbohydrates at early and late exponential growth phases. *Carbohydrate Research*, 341(12): 2090-2097.

Smith, A., M. S. Jackson and H. Kennedy (2004). Antimicrobial susceptibility of viridans group streptococcal blood isolates to eight antimicrobial agents. *Scand Journal of Infectious Diseases*, 36 (4): 259-63.

Sohn, W., B. A. Burt and M. R. Sowers (2006).Carbonated soft drinks and dental caries in the primary dentition. *Journal of Dental Research*, 85: 262.

Song, L., P. Sudhakar, W. Wang, G.Conrads, A. Brock and J. Sun *et al*. (2012). A genome-wide study of two-component signal transduction systems in eight newly sequenced mutans streptococci strains. *BMC Genomics*., 13: 128.

Steiner, M., J. Villablanca, J. Kersey, N. Ramsay, R. Haake, P. Ferrieri and D. Weisdorf (1993). Viridans streptococcal shock in bone marrow transplantation patients. *American Journal of Hematology*, 42(4): 354-358.

Svensater, G., B. Sjogreen and I. R. Hamilton (2000). Multiple stress responses in *Streptococcus mutans* and the induction of general and stress-specific proteins. *Microbiology*, 146: 107-117.

Tamura, S. (2008).Effects of oral streptococci on biofilm formation by cariogenic bacteria in dual species cultures.*KokubyoGakkaiZasshi*., 75(1): 38-48.

Tamura, S., H. Yonezawa, M. Motegi, R. Nakao, S. Yoneda, H. Watanabe, T. Yamazaki and H. Senpuku (2009). Inhibiting effects of *Streptococcus salivarius* on competence-stimulating peptide-dependent biofilm formation by *Streptococcus mutans*. *Oral Microbiology and Immunology*, 24: 152-161.

Thanyasrisung, P., Komatsuzawa, H., Yoshimura, G., Fujiwara, T., Yamada, S., Kozai, K., Eto, K., Luzumi, Y. and M. Sugai (2009). Automutanlysin distupts clinical isolates of cariogenic streptococci in biofilms and planktonic cells. *Oral Microbiol Immunol.*, 24(6): 451-5.

Tamura, S., H. Yonezawa, M. Motegi, R. Nakao, S. Yoneda, H. Watanabe, T. Yamazaki and H. Senpuku (2009). Inhibiting effects of *Streptococcus salivarius* on competence-stimulating peptide-dependent biofilm formation by *Streptococcus mutans*.*Oral Microbiology and Immunology*, 24: 152-161.

Tollefson, L. and M. A. Miller (2002). Antibiotic use in food animals: controlling the human health impact. *AnimBiotechnol*., 13(1): 71-84.

Tremblay, Y. D. H. Lo, Y. H. Li, S. A. Halperin and S. F. Lee (2009). Expression of S. mutans essential two component regulatory system VicRK is pH and growth phase dependent and controlled by the LiaFSR three component regulatory system. *Microbiology*, 155(9): 2856-65.

Tunkel, A. R. and K. A. Sepkowitz (2002). Infections caused by viridans streptococci in patients with neutropenia. *Clinical Infectious Diseases*, 34(11): 1524-1529.

Volk, W. A., D. C. Beenjamin, R. J. Kandeer and J. T. Parson (1991). *Essential Medical Microbiology*, 4th Edition, pp. 353-374, 564-566.

Vriesema, A. J. M., R. Brinkman, J. Kok, J. Dankert and S. A. J. Zaat (2000). Broad-host-range shuttle vectors for screening of regulated promoter activity in viridans group streptococci: Isolation of a pH regulated promoter. *Applied Enviornemntal Microbiology*, 66(2): 535-542.

Whiley, R. A., D. Beighton, T. G.Winstanley, H. Y. Fraser and J. M. Hardie (1992). *Streptococcus intermedius*, *Streptococcusconstellatus*, and *Streptococcus anginosus* (the *Streptococcus milleri* group): Association with Different Body Sites and Clinical Infections. *Journal of Clinical Microbiology*, 30(1): 243-244.

Woodward, E. J. (1999). The effects of traditionally prepared herbal decoctions on the growth of indigenous oral microbiota.*The Journal of Student Research*, 3(1).

Wu, C. D. (2009). Grape products and oral health. *The Journal of Nutrition*, 1818S-1823S.

Xue, X., J. Tomasch, H. Sztajerand I. Wagner-Do¨ bler(2010). The delta subunit of RNA polymerase, RpoE, is a global modulator of *Streptococcus mutans* environmental adaptation. *J Bacteriol*, 192: 5081-5092.

Zhang, G. L. and Q. J. Qian (2004). Massive ascites due to *Streptococcus acidominimus*: report of one case. *Di Yi Jun Yi Da Xue Xue Bao*., 24(5): 545.

(*Accepted for publication November 2022*)