Review Article

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Dental Caries Vaccine – A Possible Option?

Dentistry Section

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ABSTRACT

Dental caries is an irreversible microbial disease of the calcified tissues of the teeth and it has a multifactorial origin. In India, the dental caries prevalence in 35-44 year olds was reported to be 80-95% in a DCI survey. Among the elderly in the 65-74 years age group, the DCI survey reported the caries prevalence to be about 70%, while the present survey reported it to be 51- 95% in various states. Surveys which were done on school children in India showed a carie prevalence of approximately 58%.Among the U.S. population, a survey showed an incidence of 93.8% in adults with either past or present coronal caries and an incidence of 45.3% in children 23. In countries like Brazil and China, it is reaching epidemic proportions. Thus, more effective public-health measures are needed to combat dental caries. *Mutans streptococci* is one of the main microorganisms which are associated with the aetiology of dental caries. Preclinical studies of immunological interventions have shown that the disease can be interrupted. Clinical trials have indicated that a mucosal immune response to *Streptococcus mutans* crucial antigens can influence the pathogenesis of dental caries. The dental caries vaccine, when it is used in appropriate individuals at the appropriate time, can reduce the reemergence of the disease.

Key Words: Immunity, *Streptococci*, Survey

INTRODUCTION

Dental caries is an irreversible microbial disease of the calcified tissues of the teeth, which is characterized by demineralization of the inorganic portion and destruction of the organic substance of the tooth, which often leads to cavitation [1]. Dental caries is a multifactorial disease, which is caused by host, agent, and environmental factors. Immunity refers to the resistance which is exhibited by the host towards injuries which are caused by microorganisms and their products. Immunity can be innate immunity or acquired immunity. Acquired immunity can be actively acquired or passively acquired. IgA is secreted in saliva. The immunization against dental caries was achieved to a certain extent experimentally through Ag I/II12, the glucan binding domain of *S. mutans* GTS.B11 and more.

CLASSIFICATION OF DENTAL CARIES [1]

CLASS I	a) Pit and fissure caries on the occlusal surface of Premolars and molars b) Facial or lingual pits of molars c) Lingual pits of maxillary incisors
CLASS-II	Proximal surfaces of premolars and molars involving 2 or more surfaces
CLASS-III	Proximal surfaces of incisors and canines
CLASS-IV	Proximal surfaces of incisors and canines and also the incisal angle
CLASS-V	a)Gingival 1/3of facial surfaces of teeth b)Gingival 1/3 of lingual surfaces of teeth
CLASS-VI	a)Incisal edges of anterior teeth b)Cuspal tips of posterior teeth

[Table/Fig-1]: Black's Classification Of Dental Caries

DISCUSSION

The Microbial Aspect of Dental Caries

A) Microbes and their Characteristics

S.mutans is the primary aetiologic agent of this disease. *S. sobrinus* and lactobacilli are also implicated in this disease. The period which is called" window of the infectivity" which is between the middle of the second year and the end of the third year of life, shows more *S. mutans* colonization in children [2].

B) *Streptococcus mutans* and the local immunity

The oral immune system undergoes a rapid development with the secretory IgA antibody being secreted in the saliva at 1month of age. Within weeks of the initial exposure to *S. mutans*, the mucosal Ig A is secreted. The saliva of infants contains the Ig M and the IgA1 isotypes in the first month of life and by six to nine months of age, the adult like distribution of the salivary IgA1 and IgA2 subclasses appear.

C) The adherence of *S.mutans* and plaque formation

The initial attachment of *S mutans* to the tooth is through the interaction of the bacterial protein with lecithin in the dental pellicle. The Streptococcal adhesins [antigen I/II or PAC] in *S.mutans* bind to the tooth pellicle and then secrete glucosyl transferases [GTF] which help in the accumulation of more *S.mutans* through an interaction with the bacterial cell associated glucan-binding proteins. Then, they release lactic acid by their metabolisms, which demineralize the enamel, thus causing dental caries.

Glucosyl Transferase, Adhesion and Glucan-Binding Protein

Immune interventions can be undertaken at various stages of the caries pathogenesis. One is clearing the microorganisms in

the salivary phase before their colonization, which also can be done by enhancing the antimicrobial activity of the salivary IgA antibody. The receptors which are necessary for their colonization (eg. Adhesins) or accumulation (eg. Glucan-binding domains of GBPs and FTF) can be blocked. Inactivation of the GTF enzymes can be done.

ADHESINS

The adhesins from *S. mutans* [antigen I/II, PAc or P1] and *Streptococcus sobrinus* [SpaA or Pag] have significant sequence homologies [66%]. Crowley and colleagues (1993) and Nakai and co workers (1993) described that the alanine rich region could bind to the salivary component in experimental tooth pellicles. Lehner Kelly and coworkers (1994, 1995) suggested that it was through the proline-rich central portion. Active immunization with the use of an intact antigen I/II 12 or passive immunization with the use of a monoclonal 16 or a transgenic antibody 17 to the putative salivary binding domain epitopes within this component can protect rodents, primates or humans from the dental caries which is caused by *S mutans*. Protection is achieved by an antibody blockade of the initial colonization events or an antibody mediated agglutination and clearing of adhesin bearing bacteria from the saliva.

Glucosyl Transferases [GTF]

Glucosyl transferase are synthesized by *S.mutans* and S.sobrinus. The GTF activity is achieved through the glucan-binding function. GTF B 22, GTF C18 and GTF D 9 are the genes which are responsible for the glucan synthesis. Active immunization can be achieved with GTFs of *S. mutans* or *S. sobrinus*, since it induced protective immune responses in rodent models after an infection with *S. mutans* [3].

Glucan-Binding Proteins

S.mutans have cell-wall associated glucan binding proteins [Gbp]. Many proteins such as 35 GbpA 19, GbpB31 and GbpC20. have glucan-binding activities. Gbp A has a C-terminal region with [4] repeating units and it represents the glucan-binding domain of this protein [Haas and Banas. 2000]. The GbpB proteins have a role in the bio-film formation on plastic surfaces [5]. GbpC is non-enzymatic and it has a sequence similarity with the AgI/II adhesin family. In experimental studies, GbpB was found to induce a protective immune response [6]. This was achieved by a subcutaneous injection of GbpB in the salivary gland region 26 or a mucosal application through the intranasal route. In saliva, the IgA antibody to GbpB produces a natural induction of immunity in young children [7]. GbpA is less immunogenic and in S.sobrinus, GbpS has not been evaluated.

Secretory immunity and the Synthesis of IgA

IgA is the second most abundant class which constitutes 10-13% of the serum immunoglobulins and a half life of 6-8 days. It is the major Ig in saliva and tears. It occurs in two forms. Serum IgA is found on the mucosal surface and in secretions, it occurs as a dimer which is called secretory IgA. The dimeric IgA is synthesized by the plasma cells which are situated near the mucosal or the glandular epithelium. IgA is secreted in saliva in as early as the first month of life and within 6 months of life, an adult-like IgA formation becomes complete 39.

The Function of IgA And Its Immune Response

The IgA antibody functions by inhibiting the adherence of microorganisms to the surfaces of the mucosal cells and by covering the organisms, thereby preventing their entry into the body tissues. It promotes phagocytosis and the intracellular killing of microorganisms. Thus, by inducing Ig A formation by using the dental caries vaccine, the pathogenesis of the caries formation is interrupted and the dental caries formation is prevented.

The Prospects of Dental Caries

The immunization against dental carries should begin as early as the second year of life, as the population of this age group is under a normal risk of this infection. Both active and passive approaches have shown success in animal models and in human clinical trials. Understanding the colonization signals and the growth of cariogenic Streptococcus in dental biofilms is important for creating a refined technique to clear or lock harmful bacteria. The subunits of vaccines contain the structural elements of the Ag I\II adhesin family, GTFs or Gbp B [8].

In synthetic peptide vaccines, monoclonal antibodies are raised by an immunization with the intact Ag I\II, that reacts with the proline rich fragment and inhibits dental carries formation experimentally [9]. Monoclonal and polyclonal antibody preparations which are directed to several N-terminal GTF proteins inhibit the GTF activity [10, 11]. Synthetic peptide constricts were also used. These studies suggested that a protection could be achieved by immunization with discrete epitopes which were associated with several virulence characteristics.

The recombinant vaccine expresses a major part of the functional domains. The genetically linked, 42 kDa salivary binding receptor (SBR) of S mutants, AgI\II, with the A2 and B subunits of cholera toxin, produced a chimeric protein, which on intranasal administration, reduced the dental caries in Fischer rats [12].Conjugate vaccines are produced by conjugating bacterial polysaccharides with functionally associated proteins \peptides.

TREATMENT

The Human Application of Immunization
Active Immunization

Only few clinical trials have been performed in this field .When humans are immunized with glucosyl-transferases from *S. mutans* or *S. sobrinus*, there is a formation of the salivary Ig A antibody at modest levels. Enteric coated capsules with crude *S. mutans* GS-5 GTF antigen preparations which were contained in liposomes, orally immunized some adults [13]. A mucosal immunization with GTF influenced the re-emergence of mutan *Streptococci* in young adults after a dental prophylaxis [14, 15]. A topical administration showed a delay in the emergence of *S mutans* when GTF was applied on the lower lip.

Passive Immunization

When mouse monoclonal IgA or the transgenic plant secretory IgA/G antibody was topically applied [4, 5], recolonization of the *mutans* ***Streptococci*** did not occur at least for two years after the treatment. Monoclonal antibodies, in the secretory form, are more effective, because they have increased survival times in the oral cavity as compared to IgA [5].Young children who are not infected with *S.mutans* during the window of infectivity remain undetectably infected for several years [2, 10]. The Sinche niche in the dental biofilm was filled by other indigenous flora. Experimentally, this could be achieved with the use of the antibody to GTF or GbpB [16].

The Routes of Administration

The oral route was used earlier, but it was not effective due to the determinental effects of the stomach acidity on the antigen and as the inductive sites were far away. The intranasal route targets the nasal associated lymphoid tissues [17]. With the *S. mutans* antigen, AgI/II12 and the glucan-binding domain of *S.mutans*, GTF-B11, a protection could be demonstrated. The tonsillar vaccine can induce an IgA response. The tonsillar application of a particular antigen can induce IgA production in both the major and minor salivary glands of rabbits [18]. A labial application of GTF on the minor salivary glands resulted in a lower proportion of indigenous *Streptococci*/total *Streptococcal flora* in the whole saliva in next 6 weeks period [15]. The rectal route remotely induces salivary IgA responses to the *S.mutans* antigen such as GTF [19]. The Cholera and E.coli heat liable enterotoxins, liposomes, microparticles and macroparticles act as adjuvants and help in delivering the dental caries vaccine.

Recent studies

The *S sobrinus* recombinant enolase (rEnolase) is used as a target antigen. rEnolase plus an alum adjuvant was delivered into the oral cavity of rats. It increased the levels of salivary IgA and the IgG antibodies which were specific for this recombinant protein. These results indicated that rEnolase could be a promising and a safe candidate for testing in the trials on vaccines against dental caries in humans [20]. The suppressive effects of lozenges which contained egg yolk antibodies (immunoglobulin Y [IgY]) against the *Streptococcus mutans* cell-associated glucosyltransferase (CA-gtf) was studied in healthy young adults. The results of the study showed that the lozenges which contained anti–CA-gtf IgY could suppress the oral colonization by mutant *Streptococci* in healthy young adults. All vaccines, if they are properly manufactured and administered, seem to have no risks. The most serious risk is that the sera of some patients with rheumatic fever show a serological cross-reactivity between the heart tissue antigens and certain antigens from haemolytic *Streptococci*. In experiments which utilized antisera from rabbits which were immunized with the whole cells of *S. mutans* and with a high molecular weight protein of *S. mutans* were reported to cross react with the normal rabbit and human heart tissues. Polypeptides which are immunologically cross-reactive with the human heart tissue and myosin from rabbit skeleton muscles, are found in the cell membranes of *S. mutans* and Streptococcus ratti. The signals of the colonization and growth of carcinogenic *Streptococci* in dental biofilms may help us devise more refined and informed techniques to “lock out” those bacteria that can cause us harm. In gnotobiotic rats, the ingestion of whole *S. mutans* selectively produces S-IgA. The appearance of S-IgA correlated with a reduced incidence of the caries vaccine. The principal design in most of the experiments has been to first immunize the animals with an antigen from *S. mutans* which was incorporated in an adjuvant, as frequently as was necessary, to attain high antibody levels, and to follow this by implanting the same organism in the mouth and placing the animals on a high sucrose diet. As dental caries fulfills the criteria of an infectious disease, the possibility of preventing it by vaccination has been pursued. The rationale is that the immunization with *S. mutans* should induce an immune response, which might prevent the organism from colonizing the tooth surface, thereby preventing decay. The vaccine could be given at the same time when the vaccines against diphtheria and tetanus are given. The immunity could be boosted at intervals thereafter, to provide a life-long protection.

CONCLUSIONS

Dental caries is an irreversible microbial disease. The primary aetiologic agents for dental caries are *Streptococci* mutans, S.sobrinus and Lactobacillus. Through adhesions, *S.mutans* attaches to the dental pellicle and through the formation of GTF and then glucan, more organisms colonize and lactic acid formation is initiated, thus causing dental caries. An immune intervention can be undertaken by blocking the receptors which are necessary for the colonization of these bacteria or by inactivating GTF. Through these measures, the immunization against dental caries can be achieved . This may help greatly in improving the oral health in the developing countries.

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