PREVENTIVE MEASURES FOR BIOFILM ON DENTAL IMPLANTS - A REVIEW

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Abstract

Dental implants are fixtures with subsequent restorations in the form of crowns and bridges placed in the location of missing teeth. Its care is of utmost importance, to prevent the accumulation of a legion of bacteria to form a slimy sticky layer called a biofilm. Biofilm formation on oral implants can cause inflammation of peri implant tissues, which endangers the long-term success of osseo-integrated implants. Prevention of biofilm formation is necessary to ensure the long activity of restoration. Commonly, surface coatings, dispersal agents, heat treatment, nano structuration and bacteriophage releasing materials are used. Material topography and composition have also been studied. This review was done to raise awareness about biofilms in dental implants and to analyze and compare the previous methods of prevention and recent advances in the field of biofilm formation.

Keywords: *Biofilm, dental implants, titanium, Peri implant tissues, Periodontitis*

INTRODUCTION

Despite improvements in dental care, millions of people around the world suffer tooth loss. Mostly due to tooth decay, periodontal disease or injury. For many years, the only treatment options available for people with missing teeth were bridges and dentures. But, today, dental implants are available[(1)]. Dental implants are artificial tooth roots that are placed into the jaw to hold a replacement tooth orbridge[(2)]. It improves the appearance, improves speech, improves comfort, is your eating, improves self-esteem and improves overall health[(3)]. They are also very durable and very convenient for patients. Success rates of dental implants vary, depending on where in the jaw the implants areplaced, but, in general, dental implants have a success rate of up to 98%. With proper care, implants can last for life time. They require the same care as real teeth, including brushing, flossing, rinsing within an antibacterial mouthwash, and regular dental checkup. Most implants are made of titanium, which allows the host to integrate very easily. Failure of dental implant is mainly due to multiple complications arising due to a main cause that is biofilm formation[(4)].

Biofilm is a thin, slimy layer of bacteria that adheres to surfaces in the mouth such as tongue, gums and teeth. When brushing, flossing and rinsing habits are lacking, the biofilm builds up and develops into dental plaque[(5)]. Microorganisms form an attachment to the surface of the tooth, by secreting a slimy, glue-like substance. Excess biofilm formation causes plaque accumulation which may cause inflammation reactions around the

implants, leading to implant failure[6]. Most commonly and predominantly, streptococci are colonizing microorganisms, to which Prevotella species, fusobacterium, Capnocytophaga, bind and involve periodontal infections. Most commonly streptococcus mutans, streptococcus sanguini, streptococcus salivarius and Streptococcus anginosis [7].

BIOFILM FORMATION

An oral biofilm is formed when bacteria undergo a cycle of attachment to the surface of a tooth, maturation, colonization, release and spread to other surfaces[8]. The initial attachment of bacterial cells is the critical stage for biofilm formation. The excreted polymeric compounds (EPS) Matrix is important for the protection, adhesion, stabilization and nutrients within the biofilm(9). Dental implants are a consequence of periodontal disease, due to periodontal pockets, which are a reservoir of microbial pathogens. These bacteria may proliferate to form a biofilm layer. The surface properties of the implant, including roughness, may affect bacterial adhesion and contribute to biofilm formation[10]. Surface and socket of dental implants are exposed after surgery, increasing the possibility of biofilm formation. The contamination, debridement and removal of implant threads are a way of removing biofilm on dental implants [11].

The roughness of the intra-oral surfaces has a major impact on the initial adhesion and the retention of microorganisms, and if the roughness were sub-gingival, the retention of the microorganisms would be more. Studies clearly revealed that the initial adherence and colonization on the tooth enamel started where surface irregularities were present[12]. These surface irregularities include cracks, grooves and abrasion defects. The colonization of bacteria then spreads out from these irregularities to other areas of teeth. Surfaces in the oral cavity such as the dorsum of the tongue roughened by presence of papilla and the desquamating epithelium of the mucosa harbors other surfaces for the adhesion of bacteria[13]. Along with these surfaces, the enamel surface of gingival crevices and the tonsils are also believed to be the sites where bacterial adhesion occurs. Microorganisms as stated by the authors are specifically present at these sites, but they are believed to exist on all the hard and soft tissues of the oral cavity. Dental implants with rough surfaces provide microorganisms with a surface to attach with[14]. The added friction helps bacteria to adhere strongly and hence reproduce rapidly. In comparison, dental implants having smooth and shiny surfaces have less surface area and low friction, and so attachment of bacteria is difficult. Dental implants having rough surfaces thus are a predisposing factor to the formation and maintenance of biofilms[15].

PREVENTION OF BIOFILM FORMATION

One main factor affecting biofilm formation is considered to be the surface of dental implant(16). A study has shown that titanium implants with smooth surfaces provide less surface area and friction for attachment and addition of microbes in comparison to implants with rough surfaces. Rough surfaces have very large surface areas. However, most research disproves this point [17]. Previous researches also looked at factors such as material stiffness, and claimed that denser and hence stiffer materials are more prone to biofilm formation, but this theory is largely overlooked nowadays[18]. Most pathogenic bacteria are mesophilic and thrive at temperatures between 33° and 41° Celsius. Elevated temperatures inhibit bacterial proliferation and mobility, which in turn, can increase autolysis and cell wall damage. Hence, heat treatment is a very common way of arresting bacterial colonization[19]. The material of dental implant also plays a role in biofilm formation. A study showed that PMMA (polymethyl methacrylate) is the most susceptible to bacterial colonization, followed by stainless steel and in titanium(20). Further studies have demonstrated that PMMA is capable of hosting biofilms that can cause acute, chronic and delayed onset infections[21]. Implants are often coated with substances that are antibacterial or antimicrobial in nature. They are usually hydroxyapatite, sprayed titanium and titanium alloy. However these have shown little difference in microorganism concentration[22]. Dispersal agents like alpha amino acids,cis-2-deconic acid, nitric oxide and some naturally occurring peptides and enzymes have been identified which inhibit growth of bacteria in biofilm(23). Another method is micro structuration or nano structuration, which allows the surface to encapsulate air in between, creating an air cushion which reduces the water addition onto the surface, thus preventing the addition of microorganisms and other fouling molecules[24]. Recently, scientists are also studying bacteriophage releasing materials, which target certain bacteria[25]. These are difficult to incorporate in the oral cavity, as other useful bacteria present, might also get killed. Therefore, these methods are not always used very often[26].

The coating of dental implants with bioactive molecules is a recent approach to modify pristine Ti biochemical properties and hinder bacterial colonization of implant surfaces[27]. Implant coatings are essentially based on two strategies: drugs or biomolecules are mixed within the bulk of the device and released through diffusion or degradation of the matrix, or they can be grafted to the implant surface[28]. With the purpose of preventing implant infection, these strategies have been implemented using different drugs and molecules or constructs to obtain anti-bio-adhesive coatings, antimicrobial coatings (e.g., Vancomycin, Ag, Zn) or coatings (e.g., calcium phosphate, polylactic acid, chitosan) with controlled release of antimicrobial agents[29].

PRIOR RESEARCH

Several types of coatings have been investigated, including PEG (poly ethylene glycol), modified chitosan, antibiotic loaded polymer coating, zwitterionic coatings, formulations of silver, immobilised enzymes and various combinations of these are still being developed(30). Zirconia implant surfaces, in previous studies, showed statistically significant reduction in human plaque biofilm formation after 72 hours of incubation in an experimental anaerobic floor chamber model compared with titanium implants offices using dental ceramics as the implant material[31].

Coatings previously tested on dental implants:

A.**Bacteriostatic Materials**

1. Poly-Cations and Polysaccharide Coatings - Chitosan is a natural biodegradable and biocompatible polysaccharide derived by deacetylation of chitin (polysaccharide found in the exoskeleton of crustaceans and insects). It has been widely investigated as a natural biomaterial for many biomedical applications due to its biocompatibility, biodegradability, antimicrobial properties, and functionality. Chitosan is non-toxic, so it is used in medical applications such as antimicrobial and wound healing biomaterials. It is also used as a chelating agent due to its ability to bind with cholesterol, fats, proteins and metal ions. Regarding chitosan and alginate coatings, they are thought to have a surface charge and hydrophilicity that could be biostatic, hence maintaining the antibacterial ability after the complete release of minocycline. This also resulted in an improved sustainability of minocycline release[32]. Thus, the antibacterial activity was improved. This type of strategy could inhibit the immediate colonization of bacteria onto implant surfaces in the course of dental implant surgery, and thereby prevent and reduce the occurrence of peri-implantitis [33].

2. Polymers - Poly(N-isopropylacrylamide)

(polyNIPAM) is one of the most studied and widely- used environmentally sensitive (smart) polymers for controlling wettability of surfaces. PolyNIPAM is a thermoresponsive polymer that exhibits a lower critical solubility temperature (LCST) in water at 32°C. PolyNIPAM is a means of controlling bacterial attachment prevention (P. gingivalis and S. aureus). Indeed, Ti surfaces coated with polyNIPAM can detach bacteria when the temperature decreases[34].

B. **Bactericidal Materials**

1. Polymer coating - Microbial cells generally carry a negative net charge at their surface due to their membrane proteins, teichoic acids of Gram-positive bacteria, and negatively charged phospholipids at the outer membrane of Gram-negative bacteria[35]. This way, polycations are attracted and if they have a

proportionate amphiphilic character, they are able to disrupt the outer as well as the cytoplasmic membrane and enable lysis of the cell resulting in cell death [36].

2. Antimicrobial peptides - AMPs display a wide range of activity being antiviral, antibacterial, antiparasitic and antifungal. They can be derived from human as well as animal sources. Human derived peptides are advantageous over antibiotics since they are biocompatible, display low host cytotoxicity and possess broad spectral activity. Peptides can be tethered to titanium to confer an antimicrobial effect to inhibit biofilm formation. Tet213, a cationic peptide, bound to titanium has demonstrated bactericidal action on s.aureus and p.aeruginosa [37].

3. Ion implanted surfaces- Elements such as fluorine (F), zinc (Zn) calcium (Ca), chlorine (Cl), iodine (I), copper (Cu), cerium (Ce) or selenium (Se) may be incorporated into titanium or hydroxyapatite coatings by anodic oxidation of the corresponding ions. The bactericidal activity of these ions seems to depend on their gradual release from specimens into surrounding tissues[38].

4. Photo activated bio-active titanium - In an in vitro study under static and dynamic conditions, UVA illumination prior to bacterial colonization induced a reduction in adhesion rates and a significant decrease in the adhesion strength of S. epidermidis and S. aureus, without altering biocompatibility. UVA irradiation results in a super hydrophilic surface (contact angle less than 20 degrees) which oxidises adsorbed organic impurities and produces reactive oxygen species thereby rendering the titanium surface highly antimicrobial. Hydroxyl radicals (HO-) and H2O2 are produced which can destroy the bacterial cell membrane[39].

5. Nanomaterials - Copper, zinc, magnesium and especially silver and gold NPs display antimicrobial activity and are therefore possible candidate molecules for antimicrobial implant surface modifications[40].

6. Citric acid - CA has demonstrated the greatest decontamination capacity with respect to both the killing and the removal of biofilm cells[41]. Moreover, the combination of effects is clinically desirable because it promotes biocompatibility and healing around a previously contaminated implant surface[42].

7. Antibiotics - One of the approaches to avoid bacterial infection on implants, and hence prevent biofouling, is to cover them with the coatings that can release antibiotics or antiseptics in the local niche. Such coatings can be prepared either by soaking the carrier material in a solution containing antibiotics/antiseptics or by directly impregnating antibiotics/antiseptics onto the coating material[43].

8. Nanostructures - Nanotechnology deals with the design, characterization and application of structures in the nanometer scale. Nanostructured surfaces are of great interest in dental implantology since they have been observed to confer enhanced biologic integration as well as antimicrobial properties on titanium. It was observed that TiO2 nanotubes display antibacterial properties. A combination of heat treatment and nanostructured patterning through anodization has been observed to reduce the number of dead and live bacterial cells in laboratory studies. Anatase surfaces of titanium was also found to be antimicrobial when compared with rutile surfaces [43].

9. Graphene coatings - Graphene is a one-dimensional single atom thick layer of carbon atoms connected in a hexagonal configuration. Dry and wet transfer techniques have been used to coat graphene on titanium surfaces. Thin films of graphene with silver nanoparticles have been observed to inhibit s.mutans and p.gingivalis with cell viability inversely proportional to the concentration of graphene and silver [42, 43].

10. Nitride coatings - Titanium nitride offers excellent chemical stability, corrosion resistance and enhanced hardness. The characteristic golden colour of titanium nitride aids in camouflage of the metallic fixture components under gingival tissue, leading to better aesthetics. Nitride coatings have exhibited antibacterial effect against s.mutans [42, 43].

RECENT ADVANCES

Previously our department has published extensive research on various aspects of prosthetic dentistry [16,44-52], this vast research experience has inspired us to research this topic. The coating of dental implants with bioactive molecules is a recent approach to modify pristine Ti biochemical properties and hinder bacterial colonization of implant surfaces. A natural biomolecule which has been shown to possess interesting biological characteristics is silk sericin[53]. Sericin has been recently revalued due to its properties when in contact with biological materials, making it usable in the pharmacological, cosmetic and biotechnological fields[54]. While some positive biological effects, as for example antioxidant or anti- inflammatory behavior, have been proven, the anti-bacterial efficacy of sericin is still debated and it is described by studies that often provide contrasting results[55]. The exact anti- biofilm inhibition mechanism of thesericin-based coating on S. aureus biofilms is not

yet completely known but it seems to be related to the damage of the cell membrane. After exposure to sericin, in fact, the integrity of the membrane is weakened and the metabolism is blocked, thereby eventually inhibiting the growth and reproduction of S. aureus[56].

LIMITATIONS

Most methods have been tested in laboratories and are yet to be tested clinically in real-time scenarios. They are difficult to incorporate and could potentially show many unintended side- effects and secondary effects. Interference by environmental factors is also a variable which cannot be predicted without clinical case studies.

CONCLUSION

Biofilm formation on oral implants can cause inflammation on Peri implant tissues, which endangers the long-term success of osseo-integrated implants. Prevention of biofilm formation is necessary to ensure the longevity of a restoration. The methods of nanostructured surfaces, photo-activated bio active implants and ion implanted surfaces show potential as viable options to be incorporated in clinical situations. However, it cannot be stated as of yet that there is any one method which can predictably prevent the formation of biofilms without any clinical testing and observation.

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