Review Article

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DENTIN BIOMODIFICATION:A REVIEW ARTICLE

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ABSTRACT:

Aim: To review the benefits of dentin biomodification and the use of various agents for root biomodification and update the current advances of root biomodification in periodontal regenerative therapy

Materials and Methods: Google scholar database is searched using “Dentin Biomodification” and “Root Biomodification on periodontal regeneration”.

Results: Based on the method of root biomodification they are categorized as mechanical and chemical. Mechanical modification involves scaling and root planning and chemical methods involves root conditioners such as citric acid, Tetracycline HCl, EDTA, Fibronectin, Laminin, Doxycycline, Minocycline, Polyacrylic acid, Phosphoric acid, Formalin etc. other agents used are enamel matrix proteins, Platelet rich plasma, Recombinant human growth factor, Hyaluronic Acid, Lasers. The use of these agents removes the smear layer that exposes the dentinal collagen and becomes chemo-attractant for periodontal fibroblast.

Conclusion: The present status suggests that root biomodification does have added advantage in periodontal regeneration.

Key words: Dentin biomodification; Root biomodifiers; periodontal regeneration; laser; root canal irrigants

INTRODUCTION:

Dentin is a complex mineralized tissue arranged in an elaborate 3-dimensional framework composed of tubules extending from the pulp to the dentin–enamel junction, intra-tubular, and peri-tubular dentin [1]. Dentin analogues are very similar to bone, where odontoblast-like cells are located within alveoli [2,3]. The diameter of tubules can vary between 2 and 4 µm. The number of dentine tubules is about 18 000 and 21000 tubules per mm^2 [4]. Fibrillar type I collagen accounts for 90% of the organic matrix, while the remaining 10% consists of non-collagenous proteins, such as phosphoproteins and proteoglycans [5]. Dentin undergoes modifications by physiological aging and disease processes to produce different forms of dentin [6]. This process affects the biomechanics and biochemistry of the tissue. Dentin does not share the same ability to remodel. This limits site regenerative therapies. An advantage of dentin is the presence of collagen which acts as a scaffold that provides a backbone that is cell-free for tissue repair and regeneration. Biomodification of dentin has been investigated as a biomimetic strategy therapy to

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mechanically strengthen the existing collagen network and also control biodegradation rates of extracellular matrix (ECM) components [7]. Electronic microscopic examination of gingival fibers which originates in the cementum beneath the junctional epithelium results in destruction mostly seen from the cemental surface than at the root surface. After the destruction of gingival fibers, a more extensive breakdown involving the cemental hard tissue can be observed. Ultrastructural changes seen in the hard tissue in periodontally affected teeth are a reduction in the number and size of the mineral crystals as well as the loss of collagen structure in the affected zone. Underlying dentin gets exposed by cemental resorption .dentinal tubules are exposed to the oral environment where bacteria may get demonstrated in intratubular sites. The surface of cementum in relationship to normal tissue is characterized by projections above the mineralized plane. These projections represent minute pyramids of mineralization of collagen fibers. The root surface associated with periodontal disease is covered with a cuticular material [8]. Exposed root surface on periodontally affected teeth prevents cell attachment and fiber development. Alterations are contamination of root surface by bacteria or their endotoxins, alterations in mineral density and composition and reduced chemotactic stimuli for migration of cells [9].

HISTORICAL BACKGROUND

The concept of the use of acid demineralization in periodontal therapy was first introduced in the 1800s as a substitute for calculus removal. The use of acid as an adjunct to scaling and calculus removal was reported in the New York Dental Records in 1846 [10]. In 1833, Marshall et al. reported a case of pocket eradication after the use of aromatic sulfuric acid. In the 1890s Younger and Stewart described the use of acids with mechanical removal of calculus and cementum and they found that there was hypermineralization of diseased roots with obliteration of lacunae of cellular cementum by calcific deposits[11]. According to Urist et al.,1966 they conducted a series of experiments on allogenic dentin matrix following total demineralization with 0.6N HCl showed the ability to induce the formation of new bone or cementum on the implant surface[12]. Urist et al. encouraged Register et al in 1973 to perform a study on the use of acid on root surfaces and found that there was a new attachment, cementogenesis, and osteogenesis that took place[13]. Polson and M.P. Proye studied the effect of citric acid treatment of the denuded root resulted in new connective tissue attachment, and the response appeared to be dependent upon the early establishment of fibrin linkage with the root surface[14]. Terranova, et al. in 1986 have shown that the tetracycline treatment of root surface suppresses laminin-binding and epithelial cell growth and attachment[15]. Nyman, Lindhe, and Karring studied the healing process following surgical

treatment and root demineralization in monkeys with periodontal disease and reported that citric acid conditioning of the root dentin surface did not aid in cementum formation and new connective tissue attachment[16].

REGENERATION AND DENTIN BIOMODIFICATION

Scaling, root planing and flap surgery alone result in a wound that heals with long junctional epithelium. This type of healing is called repair. Guided tissue regeneration is a technique that favors cell repopulation on the root surface by periodontal ligament cells, osteoblasts the following wounding. The faster-growing soft tissue cells such as gingival connective tissue and the epithelium are inhibited[17]. To achieve new attachment on the diseased root surface it is necessary to eliminate calculus, bacterial plaque, and other cytotoxic substances on or within the root surface. Periodontally affected root surfaces are hypermineralized and contaminated with cytotoxic and other biologically active substances[18]. The non-surgical periodontal therapy consists of scaling and root planing removes deposits on the root surface as well as the removal of diseased exposed cementum. Periodontal instrumentation leaves behind a smear layer on the root surface which remains interposed between the gingival flap and root surfaces[19]. This layer is composed of microorganisms, cementum fragments, plaque, and calculus and cementum matrix ranging in thickness by 2-15micrometer. This layer prevents new connective attachment to the root following periodontal regeneration procedure. Root surface bio-modification involves the application of a week acid on the root surface which removes the smear layer from root surfaces. This exposes the collagen fibers, produces a zone of demineralization and opens and enlarges the dentinal tubules. The exposed dentinal collagens are supposed to act as a chemoattractant for periodontal fibroblast[20].

INTERFACE BETWEEN THE TOOTH AND BIOMATERIALS

Secondary caries can be a cause for the replacement of resin composite restorations [21]. In resin dentin bonded interface collagen-based dentin matrix scaffold is reinforced by resin to produce a hybrid layer that couples resin composites to the underlying mineralized dentin [22]. A hybrid layer is the weakest link at the adhesive interface bond [23]. Altered forms of the dentin show a negative impact on the bonding ability [24]. Different dentin biomodification strategies have enhanced the properties of the dentin matrix. Riboflavin agents show an increase in collagen degradation, an increase in mechanical properties and an increase in mechanical stability [25]. Glutaraldehyde is the most widely used agent in increasing the stiffness of demineralized dentin[26]. Riboflavin/UVA has the potential to reduce dentin matrix breakdown and enhance the long-term stability of dentin-resin interfaces [27].

ROLE OF ROOT BIOMODIFICATION

Demineralization treatment on affected root surface induces cementogenesis and connective tissue attachment by fibrin linkage and periodontal ligament fibroblast migration. Demineralized dentin surface have capacity to bind fibronectin than surfaces which are not demineralised [28]. Tufting of dentin and cementum matrix following demineralization will lead to increase in total surface area of the collagen and total number of binding sites for various proteins or growth factors. They also play an important role in wound healing and regeneration [29]. Root surface modifiers can facilitate effective removal of smear layer resulting from instrumentation, and cementum bound endotoxin. The apical growth of epithelium is inhibited followed by early cementogenesis [30]. These agents help enhance binding matrix proteins to dentinal collagen matrix and stimulate fibroblast attachment and growth [15].

METHODS OF ROOT CONDITIONING

Mechanical methods: it involves scaling and root planing. This procedure helps to remove cementum, softened dentin, smoothening of surface irregularities. Scaling and root planning will not make the root surface disease-free since it does not completely remove contaminated cementum, especially in apical areas. A smear layer will cover the instrumented surface. Thus alternative approaches have been introduced to overcome the limitations [31]. Chemical methods: root surfaces affected by periodontal disease undergo structural and chemical changes due to the effects of cytotoxic and other biologically active substances from periodontal pathogens [32]. Due to the presence of bacterial cytotoxins, it prevents the reattachment of gingival and periodontal cells and cannot be reversed by conventional scaling and root planing procedures alone as the infection is not completely eliminated due to production of smear layer [33]. Because of the presence of the smear layer, it prevents reattachment of the periodontal connective tissue, and surface demineralization is done to recreate a substrate for periodontal reattachement [34]. Hence chemical conditioning agents are often used to help remove the smear layer derived from bacterial products [35].

The root surface biomodification agents are broadly classified into the following categories

1.Root conditioners- Citric acid, Tetracycline HCl, EDTA, Fibronectin, Laminin, Doxycycline, Minocycline, Polyacrylic acid, Phosphoric acid, Formaline, Chlorhexidine, Hydrogen peroxide, CetylPyridium chloride, and sodium-n-lauroyl sarcosine, Cohnns factor, Bile salts, and plasma fractions

2. Enamel matrix proteins
3. Platelet-rich plasma
4. Recombinant human growth factors
5. Hyaluronic Acid
6. Lasers

ROOT SURFACE CONDITIONERS

Citric Acid:

Smear layer removal by Register in 1973 has been studied extensively. It is essential to human metabolism and is

found in many foods. It has been used in the form of citrates as anticoagulants. Endogenous citric acid from the metabolic acid cycle has been associated with the solubility of bone mineral during bone resorption. Citric acid has enhanced features thought to be relevant in the regeneration of periodontal tissues. It helps to expose the collagen, induces mesenchymal cell differentiation, extracts endotoxins and other toxic products, accelerates cementogenesis and widens dentinal tubules. Citric acid demineralization helps in the new attachment or reattachment and regeneration. It contains two or more groups in its molecule which can combine with calcium and act as the chelating agents. Citric acid acts on dentinal hydroxyapatite by releasing hydrogen ions which demineralize the crystalline structure [36]. It helps in clot stabilization on dentin surface than tetracycline, EDTA, sodium citrate or saline solution [37]. A study conducted by Register reported the reattachment of collagen fibers to previously denuded root surfaces following treatment with citric acid and application time of 2-3minutes [38]. This study demonstrated that the application of citric acid for 3 minutes using a rubbing pressure on the surface results in complete removal of the smear layer and exposure of dentinal tubules [39]. A study was conducted to evaluate the regeneration following the application of citric acid to root surfaces. The ph of the acid ranged between 1-3 and the duration of application left on for 2-5minutes. The results showed connective tissue regeneration [40]. Drawbacks associated with the citric acid application include the formation of an extremely acidic environment in the surrounding tissues, its low ph that induces cytotoxic effects when in direct contact with periodontal cells [41].

Tetracycline hydrochloride:

They are broad-spectrum antibiotics that are effective in controlling periodontal pathogens. They are derivatives of polycyclic naphthalene carboxamide. Tetracyclines have a low ph in concentrated solution and can act as a calcium chelator resulting in demineralization. They also possess anti-bacterial properties. It has been suggested that on application tetracycline drug was released in a biologically active concentration for 48hours and upto14 days [42]. They are used at a concentration of 0.5% at a ph of 3.2 and applied for 5 minutes. Properties of the tetracycline hydrochloride are: Increases fibronectin-binding which stimulates fibroblast attachment and growth, Smear layer removal, exposure of dentin tubules or collagen fibers, Endothelial cell growth factor binding to dentin, stimulating periodontal ligament cell proliferation/migration, Absorbs to enamel and dentin, acts as an antimicrobial local delivery system, Collagenolytic enzyme inhibition preventing bone resorption.

EDTA (EthylenediamineTetracetic acid):

Chelating agents such as EDTA working at neutral ph helps to preserve the

intergrity of exposed collagen fiber, early cell colonization and wound healing.neutral EDTA reduces the probability that the soft tissues of the periodontium will be damaged. It has also been found that a ph not close to neutral inhibits periodontal ligament fibroblast. Various concentrations of EDTA has been used in studies ranging 12-24%, neutral ph for 30 seconds – 3 minutes aiming to remove the smear layer and widen the dentinal tubules [43]. However the initial clot adhesion is limited with its use [44]. Use of acidic agents to demineralize the root surface had a drawback of adversely affecting the surrounding tissues. So a chemical agent that could remove the smear layer and demineralize the tooth surface at neutral ph was required. It exerts demineralizing effect through chelating divalent cations at neutral ph. Studies have shown that application of 18% EDTA on root surface improves fibroblast attachment and migration on the root surface and also facilitate oriented fiber attachment system between demineralized surfaces. Studies were conducted on the use of 24% EDTA and were applied to the root surfaces for 23 minutes showed no difference in probing depth, clinical attachment level and probing bone levels between EDTA treatment and control root surfaces [45].

Fibronectin;

It is a high molecular weight glycoprotein that is found in its extracellular tissue and is the main component that holds the clot together. It performs several functions that foster the reattachment of periodontal tissues: Promotes mesenchymal cell adhesion, chemotaxis, and growth, stimulates the coronal growth of cells from the periodontal ligament that is responsible for new attachment, favors the growth and attachment of fibroblast over the epithelial cells to the root surface, Speeds the linkage process by being chemoattractive to fibroblast and stabilizing the clot between the exposed root surface collagen and new fibers within the tissues.

Laminin

It is a glycoprotein of high molecular weight. It is capable of adhering to different substrates. It helps in the direct movement of different cell types. Studies have shown that laminin promotes gingival epithelial chemotaxis and gingival fibroblast migration. Terranova et al have demonstrated that laminin promotes epithelial cell adhesion and growth to tetracycline and glycoprotein conditioned surfaces. It is the most abundant component of basement membranes[46].

Chlorhexidine :

A study performed on the effects of postoperative use of chlorhexidine on the regeneration of bifurcation defects. On the application of chlorhexidine to the root surfaces during surgical procedures of bifurcation defects resulted in an increase in bone height but not the level of connective tissue attachment [47].

Polyacrylic acid;

It is a weak acid and has been used as a root conditioning agent. Its acid etching

property helps to remove the smear layer from the root surface. A study conducted on the comparison of polyacrylic acid and citric acid showed that polyacrylic acid applied for 20 seconds demonstrated greater connective tissue attachment to root surface when compared to citric acid applied for 3 minutes [48].

Recombinant human growth factors:

The application of growth factors for periodontal regeneration is a major focus of the research presented. Different growth factors which are believed to contribute to periodontal regeneration include the platelet-derived growth factor (PDGF), insulin-like growth factor (IGF), transforming growth factor (TGF), epidermal growth factor (EGF), fibroblast growth factor (FGF), and bone morphogenetic protein (BMP). Growth factors promote the proliferation of fibroblasts from the periodontal ligament and bone formation. On growth factor application there is improved cellular response103. Rubins et al. conducted a study using recombinant human platelet-derived growth factor with CTGs for the treatment of Miller Class I or II gingival recession defects and observed keratinized tissue gain, better root coverage and better wound healing at 6months postoperative follow-up[49].

Lasers:

The number of commercially available laser systems is limited to some infrared laser namely: Er: YAG lasers, Nd: YAG and CO2. A study concluded that the use of CO2 and Er: YAG laser at the determined power settings can treat dentin hypersensitivity and reduce its symptoms. Er: YAG laser has a greater effect on tubular occlusion with less thermal changes and acts as a useful conditioning tool [46]. The use of Nd: YAG laser showed an alteration in the periodontal pocket epithelium with the presence or absence of clinical inflammation 116. A study conducted by Bhushan et al on the effect of Er: YAG laser on root surface using a scanning electron microscope to determine the laser's ability to remove the lipopolysaccharides using infrared spectrometry. The results showed that Er: YAG laser could remove 83% of lipopolysaccharides from the root surface which suggests that Er: YAG laser is a useful root conditioner [50,51,52].

RECENT ADVANCES

PRF has the capacity to conserve open wounds and improve healing due to its ability to attract epithelial cells and micro vascularization. Thus the application of PRF for root coverage has become increasingly popular for gingival recession treatments [53]. Injectable PRF is the liquid form of PRF. It is a bioactive agent obtained by low-speed centrifugation and has the capacity of tissue regeneration. At high concentration, PRF stimulates the secretion of several growth factors and trigger fibroblast migration [54]. I-PRF is used in regenerative treatments. One of the components that make up I-PRF is fibronectin, which is an extracellular glycoprotein with a high molecular weight (appro.440kDa) [55]. The application of fibronectin to root

surfaces improves cellular proliferation from the periodontal ligament towards the supracrestal parts. Fibronectin is used as a root surface biomodification agent in periodontal surgery. I-PRF to root surface may have a positive effect on the closure of the root surface in the context of free gingival graft operations [56].

Emdogain is a commercially available formulation of enamel matrix derivatives. The main components of Emdogain are freeze-dried enamel proteins and amelogenin fraction. Polyglycolic acid acts as a vehicle to carry these biologically active proteins. The two components of the material enamel matrix proteins and polyglycolic acid vehicle are mixed immediately before the application to make a syringable gel. At physiological pH, the amelogenins are insoluble and become soluble at low or high pH. Their solubility increases with a decrease in temperature, the pH of polyglycolic acid is below 4.5 which makes it a suitable carrier for enamel matrix protein. The flap is raised to get complete access of the root surface, the granulation tissue and tissue tags are removed completely. Hand and ultrasonic instruments are used to remove deposits from the root surface. The bleeding is controlled. The root surface is conditioned with root biomodification agents and the smear layer is removed to expose collagenous matrix of dentin and cementum, the area is rinsed with saline and Emdogain gel is applied to the root surface completely, the mucoperiosteal flap is raised and sutured to obtain primary closure [57].

CONCLUSION:

The rationale for root surface biomodification is to remove the smear layer on the root surface, uncover and widen the dentin tubules, and unmask the dentin collagen matrix. Root surface biomodification is combined with guided tissue regeneration and bone grafting to achieve better results. Application of Growth factors provides an environment for regeneration. Dentin biomodification is a bio-inspired strategy to improve the properties of the dentin matrix, it is important to consider the limitations of a wide variety of available agents and the strategies used.

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