Matrix Metalloproteinases (Mmp) in Restorative Dentistry and Endodontics

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Matrix metalloproteinases (MMPs) seem to play a dual role in dentistry. While several MMPs have an important role to play in developmental defects of teeth and in caries, some MMPs also seem to have a defensive role. The main organic component of tooth structure is collagen and MMPs that degrade collagen and the extra cellular matrix have been implicated in progression of dental caries. MMPs have also been shown to be active in pulpitis and studies have shown that they can be used as diagnostic markers of pulpal inflammation. This paper reviews the role of MMPs in restorative dentistry and endodontics.

Key words: Matrix metalloproteinase; collagen; dental caries; dentinogenesis; pulpitis; diagnostic marker

INTRODUCTION

Matrix metalloproteinases (MMP) are a family of endopeptidases which are capable of degrading almost all extra cellular matrix proteins, including different collagens in their native and denatured forms. There are several types of MMPs classified based on genetic configuration: MMP-1 (Intestitial collagenase), MMP-2 (Gelatinase – A, 72kDa gelatinase), MMP-3 (Stromelysin 1), MMP-7 (Matrilysin PUMP 1), MMP-8 (Neutrophil collagenase), MMP-9 (Gelatinase B, 92 kDa Gelatinase), MMP-10 (Stromelysin 2), MMP-11 (Stromelysin 3), MMP-14 (MT 1-MMP), MMP-15 (MT 2-MMP), MMP-16 (MT 3-MMP), MMP-17 (MT 4-MMP), MMP-18 (Collagenase 4, xcol4, xenopus collagenase), MMP – 19 (RASI -1, Stromelysin-4), MMP 20 (Enamelysin), MMP-21 (X-MMP), MMP-23A (CA-MMP), MMP-23B, MMP-24 (MT5-MMP), MMP 25 (MT-MMP), MMP26 (Matrilysin-2, endometase), MMP 27 (MMP-22, C-MMP), MMP-28 (Epilysin) .[1,2]

The MMPs are secreted as proenzymes (zymogens) and are activated via stepwise mechanisms. They can be activated by proteinases or in vitro by chemical agents, such as thiol-modifying agents, oxidized glutathione, chaotropic agents, and reactive oxygen species. Low pH and heat treatment can also lead to activation. These agents and procedures probably work through disruption of the cysteine–zinc binding [3].

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Some of these MMPs have major implications in the oral region - tooth development, dental caries, pulpal and periradicular pathoses, soft tissue lesions and periodontal pathoses. Under normal and physiological conditions, MMP activity is regulated at the level of transcription of the precursor zymogens through interactions with specific extracellular matrix components and inhibition by endogenous inhibitors [tissue inhibitor of metalloproteinases (TIMPs) [4]. This review discusses the role of MMPs in the development and pathological conditions of the dental hard tissue ie., teeth.

Tooth development - Role of MMPs in health and disease

MMPs play several important functional roles in dental hard tissues. During the process of tooth formation and development, the components of the dentine organic matrix are secreted, processed and organized to form the predentin. This is an unmineralized layer of dentin. During the process of organization, some matrix components are subjected to breakdown and removed. It has been reported that collagenase and gelatinase are present in dentin and predentin ie., these MMPs are required during the organizational step of dentin formation and mineralization [5,6]. Furthermore, the main organic component of dentin matrix is Type I collagen, implying that MMP-8 has a significantly important role in the process of dentin organization and modeling. Latent collagenase has also been observed in mineralized dentin further confirming its role during organic matrix formation [5].

A developmental anomaly of the dentinal part of tooth structure, termed dentinogenesis *imperfecta* is characterized typically by substantial quantities of Type III collagen [7,8]. This implies that degradation of the type III collagen during dentin matrix organization, associated with modeling of type I collagen is essential for formation of normally mineralized dentin**.** Both MMP-1 and MMP-8 have been shown to degrade type III collagen [8]**.**

A similar situation has also been shown in the case of dentin irritation by dental caries,, for example, wherein TGF-ß down regulates MMP-8, and the reparative dentin formed has poorly organized organic matrix with type III collagen [9], in contrast to gingival cells, TGF-ß up-regulates MMP - 9 and down-regulates MMP-8 in odontoblasts [10].

In dentinogenesis, odontoblasts secrete dentin organic matrix, which, after organization in the predentin area, is mineralized. The fact that the onset of MMP-20 expression in odontoblasts coincides with the early onset of predentin synthesis and analysis of our data on the presence of dentin-bound MMP-20 suggests that MMP-20 may be involved in organic matrix organisation during dentin formation. In dentin matrix organization, the substrates for MMP-20 remain unknown, but *in vitro* it degrades casein, gelatin, fibronectin, type VI collagen, tenascin – C and laminins 1 and 5 [11-14].

Matrix metalloproteinases in dental caries

Dental caries is characterized by demineralization of the inorganic portion of the tooth, and disintegration of the organic tissues of the tooth. The major organic component of dentin is Type I collagen. The dentin extracellular matrix can be compromised by proteolytic degradation, which happens in the presence of collagenases (MMP-1, MMP-8, MMP-13) and gelatinases (MMP-2, MMP-9) [15,16].

Furthermore, it is important to note the role of MMP-20 (enamelysin) in the progression of caries. MMP-20 does not cleave Type I or Type II collagen as is evident from in vitro work Hence, this should be attributed to dentin bound MMP-20, which is host MMP. Dentin bound MMP-20 probably contributes to the early alteration in non-collagenous organic matrix during caries progression in dentin rather than degradation of collagenous matrix per se. Also, demineralization or degradation of demineralized organic matrix is required for MMP-20 released from dentin which would therefore occur during dentin caries progression [17].

Dentin bound MMPs also seem to have a defensive role in dentin caries progression by lieu of releasing growth factors. These dentin bound growth factors help in the upregulation of dentin-pulp complex defensive reactions under caries lesions. Nevertheless, the role of MMP-20 in this process is poorly understood. In the dentinal fluid, MMP-20 is present. Immaterial of any irritation to dentin, there is constant fluid flow within the dentinal tubules, which is an odontoblast regulated process [18].

In vivo studies have shown that dentinal fluid collected from cavities contains plasma proteins. Also, immunohistochemistry studies demonstrate that the MMP 20 in dental pulp has its source as the odontoblasts and factors like external irritation (eg., caries) results in secretion of MMP-20 which may be involved in defense reactions which include reparative dentin formation. Nevertheless, this concept is yet to have solid proof. The activity of MMP proforms is elicited by dentin matrix protein-1, osteopontin and bone sialoprotein which are members of the SIBLING proteins family and are present in the dentin [19].

MMP-1, MMP-8 and MMP-13 are the most important human collagenases. While MMP-8 targets Type I collagen, MMP-1 targets Type III and MMP-13 prefers Type II collagen. The presence of these MMPs demonstrate the role that host derived MMPs are involved in the degradation of dentin organic matrix during caries progression. The major source for MMPs in carious dentin has been claimed to be the gingival crevicular fluid from where they are distributed to the whole saliva [20]. The dentin-pulp complex has also been stated as a possible source of MMP-2 and MMP-9. The role of MMPs in physiological conditions is further supported by the presence of a latent collagenase in demineralized human dentin matrix.

The role of MMP in collagen degradation is what has prompted several researchers to analyze the effect of MMP inhibitors on the durability of dentin bonds. These agents have been recommended as therapeutic primers after etching with phosphoric acid, to stabilize and enhance the durability of the resin-dentin interface. It has been reported that 2% chlorhexidine gluconate is capable of preventing reduction in resin-dentin bond strengths. By binding to the zinc and calcium ions, in the catalytic domain of MMP, chlorhexidine is able to inhibit MMP activity. The same has also been explained as the mechanism of action of galardin [21-23]. However, considerable controversy also exists in this aspect wherein, it has also been reported that CHX mediated preservation of resin-dentin bonds is active a few months after the endogenous collagenolytic activities are terminated. Research is also in progress wherein polymers may be used for local and sustained delivery of CHX at the resin-dentin interface.

MMPs in pulpal pathologies

Pulpal and periradicular diseases are responses of the immune system to microbiota, resulting in production of inflammatory and resorptive factors. The production of tissue type plasminogen activated factors, pro-inflammatory cytokines, substance P and vascular endothelial growth factor expression. In the case of pulpal inflammation. there is degradation of matrix protein which is mediated by endopeptidases. MMPs are able to degrade all extracellular matrix proteins. The denatured gelatins namely gelatin, laminin, elastin, fibronectin and the basement membrane zone associated collagen are degraded by MMP-2 and MMP-9 which are characterized as gelatinases. The proteolysis of extra cellular matrix seems to be a key initiating event for progression of inflammatory processes [24].

High levels of these neutrophil enzymes indicates a level of tissue breakdown that is beyond repair- a condition that is termed irreversible pulpitis. Matrix metallopeptidase 9 (MMP-9) or neutrophil gelatinase is a proteolytic enzyme (endopeptidase) produced by neutrophil granulocytes [24]. Zehnder *at al* reported that these neutrophil enzymes could be considered as markers of pulpal disease. In a classic report, they demonstrated that dentinal fluid samples from symptomatic teeth had significantly higher MMP-9 levels than those from clinically healthy counterparts [25]. This could possibly evolve into a non-invasive, chairside diagnostic marker of pulpal diseases and prognostic markers of procedures like deep caries management.

MMP family proteins elicit dual roles in the pathogenesis of inflammation, stimulating protective innate and/or adaptive immune functions, as well as tissue destruction [26]. MMP-3 elicits stimulatory effects on the proliferation and the migration of endothelial cells as well as anti-apoptotic effects on these cells *in vitro*.

The therapeutic application of MMP-3 for the injured pulp tissues of successfully induced tissue regeneration in rat incisors. A revolutionary work in this regard is one by Eba *et al*, wherein in a canine mature premolar with irreversible pulpitis, regeneration of pulp tissue was observed, with revascularization, at the end of 14 days after sealing with MMP-3 [27]. This was followed by extracellular matrix formation. However, in cases of severe inflammation. MMP-3 was unable to induce regeneration. MMP-3 selectively inhibited the expression of IL-6 which may play a role in host protection [28]. However MMP inhibitors failed to exhibit significant therapeutic efficacy in any human clinical trial.

CONCLUSION

MMPs have dual roles in dental hard tissues. Research should focus on identifying and formulating MMP inhibitors that may be used for the management of oral diseases. The application of MMPs as diagnostic and prognostic markers needs further exploration.

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