Abstract

Keratinocytes form a natural barrier that protects the body against external environment, injuries, and invasion of pathogens. The constant exposition of keratinocytes to commensals forced the development of adaptation mechanisms allowing them to tolerate the presence of microorganisms without triggering an excessive inflammatory response. Recently, MCPIP-1 protein (*monocyte chemoattractant protein-induced protein-1*) has been classified among those mechanisms. The abundant, constitutive expression of MCPIP-1 characterizes many types of epithelial cells, preventing exaggerated induction of inflammation upon contact with microorganisms. The anti-inflammatory effect of MCPIP-1 is based on its activity as endoribonuclease against transcripts of proinflammatory cytokines, including IL-1β, IL-6, IL-8, and IL-12p40. Moreover, the deubiquitinase activity of MCPIP-1 efficiently inhibits the NF-κB signaling pathway. The role of MCPIP-1 has been thoroughly described in the biology of skin keratinocytes, but nothing is known about its function in the physiology of periodontal tissue. Therefore, the presented doctoral dissertation is devoted to this issue.

The first part of the presented thesis aimed to identify the role of MCPIP-1 in the development of periodontitis of bacterial etiology and the process of alveolar bone loss. One of the pivotal species implicated in the progression of chronic periodontitis is Gram-negative bacterium *Porphyromonas gingivalis*. It produces a plethora of virulence factors that modify the host's defense response. Among them gingipains, cysteine proteases are considered the most crucial. Presented study demonstrates that gingipains are responsible for the efficient degradation of MCPIP-1 protein *in vitro*, as well as *in vivo,* in a murine model of chronic periodontitis induced by *P. gingivalis*. Depletion of MCPIP-1 promotes amplification of the inflammatory response leading to the alveolar bone loss. Analysis of the molecular basis of observed phenomenon revealed that it is related to an excessive and uncontrolled response of gingival keratinocytes selectively to lipopolysaccharide (LPS). The presented results indicate the orchestrated activity of *P. gingivalis* virulence factors. LPS recognized by TLR4 activates the signal transduction pathway triggering the expression of pro-inflammatory cytokines, while the degradation of MCPIP-1 catalyzed by gingipains ensures the stability of the transcripts of these inflammatory mediators. Furthermore, other periodontal pathogens including *Tannarella forsythia* and *Prevotella intermedia* apply a similar strategy also degrading MCPIP-1. In summary, the described phenomenon can be considered as one of the novel virulence mechanism of

„*inflammophilic bacteria*" which endure inflammation but also take advantage of it, being insensitive to the force of the host's immune system.

In the second part of the study, the role of MCPIP-1 in maintaining the homeostasis of healthy periodontal tissue was analyzed. For this purpose, a transgenic mouse lacking *Zc3h12a* gene (encoding the MCPIP-1 protein) exclusively in keratinocytes was used. Using the micro-computed tomography method, we demonstrated that mice lacking MCPIP-1 only in keratinocytes develop massive alveolar bone loss. Detailed analysis of that pathology revealed that the observed bone loss is the result of excessive expression of chemoattractants and cytokines produced by gingival keratinocytes, which in turn leads to the development of neutrophilic inflammation affecting periodontium. One of the key mediators involved in this process is IL-36α which activity is intensified by proteases produced by accumulated neutrophils. The process of local inflammation leads to a decrease in the number and activity of bone-forming osteoblasts, resulting in a progressive loss of alveolar bone. The process can be intensified by mechanical irritation of the epithelium probably *via* mechanotransduction signaling.

Taken together, presented results comprehensively demonstrate for the first time the role of MCPIP-1 protein in maintaining oral epithelium homeostasis and in the process of periodontitis development. Furthermore, the presented data complement the concept of osteoimmunology indicating that gingival keratinocytes are crucial component in alveolar bone remodeling by shaping the immune response of the oral mucosa.

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