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Interaction of hemidesmosome protein and focal contact protein in healing wound

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Objective: Keratinocytes (KCs) have two anchoring devices, hemidesmosomes (HDs) and focal contacts (FCs). From previous reports, HDs and FCs have been predicted to interact each other through laminin-332, CD151, plectin and signaling molecules. However, there have been no direct evidence of interaction between HD protein and FC protein in KCs. Therefore, we investigated the interaction between HD protein and FC protein in KCs.

Methods: To observe dynamics of HD protein and FC protein at the same time, we expressed YFP-tagged β4 integrin, and CFP-tagged α-actinin, respectively, in live HaCat cells at wound edges under several conditions and observed their dynamics by time-lapse video microscope.

Results: At the leading edge of scraped wound, FC protein assembled rapidly and regularly in the direction of the wound. Subsequently, HD protein followed and filled into the “FC protein-rich” region where FC protein disassembled. FC protein disassembled together with the appearance of HD protein and new FC protein assembled at the newly formed leading front of KCs. KCs repeated this cycle until KCs no longer moved. Under conditions that affect FCs, the HD protein dynamics became highly stable and HaCat cells ceased migration. Under conditions that affect HDs, the velocity of FC protein became more rapid and the direction of the assembly of FC became irregular. The migration of KCs was not in alignment. Under other conditions (treatment of anti-laminin-332 antibodies, transfection of CD151 siRNA, plectin siRNA, FAK and Shc siRNA), the dynamics of HD protein and FC protein were affected and the migration of KCs became irregular.

Conclusions: The interaction between HDs and FCs does occur at at least in protein level in KCs at the wound edge and that this interaction is mediated by the fine tuning of each constituent of HD, FC, signal molecules and in-between proteins.

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Inflammatory Alveolar Bone Resorption in Mouse Model of Marfan Syndrome

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Keywords: Marfan syndrome, severe periodontitis, fibrillin-1

Objective: Marfan syndrome is a systemic disorder of connective tissue, such as, skeletal, cardiovascular, and ocular systems. In addition, severe periodontitis is frequently seen in this disorder. FBN1 encoding fibrillin-1, which is a microfibrillar protein in elastic system fibers, is one of the responsible genes for this disorder. We hypothesized that abnormal fibrillin-1 expression might relate to the pathogenesis of the severe periodontitis in this disorder. In order to clarify the mechanism of the periodontitis, the mouse model of this disorder (hypomorphic Fbn-1 mouse) was challenged by Porphyromonas gingivalis (P.g.) in this study.

Methods: Hypomorph Fbn-1 mouse (6-week-old heterozygous MθA mice; n=6), which have 5 times lower Fbn-1 expression than age-matched wild-type mice (WT; n=6), were infected with P.g. after one week of the antibiotic treatment. At 2 and 8 weeks after the infection, the distance and area were measured between the cementoenamel junction and alveolar bone crest. The blood sample and were also collected from mice. The alveolar bone resorption was examined by μCT analysis (Shimadzu InspeXio SMX), and the level of TNF-α was examined by ELISA.

Results: The P.g. infection induced the alveolar bone resorption both in hypomorphic Fbn-1 and WT mice. The amount of bone resorption was significantly higher in hypomorphic Fbn-1 mice than in WT mice. This was accompanied by the higher level of TNF-α.

Conclusion: The severe periodontitis in Marfan syndrome was reproduced in mice. Findings suggest that the decreased Fbn-1 expression induces the increased level of TNF-α. The results suggest that the normal fibrillin-1 expression is indispensable for the integrity and maintenance of the periodontal tissues.