TGFbeta-Dependent Localization of MT1-MMP Regulates Epithelial Tubulogenesis in 3D Collagen

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Objectives: Epithelial tubes are essential structures in multicellular organisms. However, the fundamental mechanism underlying the formation of such structures is still unclear. Previously it was reported that MT1-MMP is essential for tubulation but its activity needs to be regulated in order to form organized structures. In this paper, we investigated regulation of MT1-MMP during epithelial tubulogenesis using a Madin-Darby Canine Kidney (MDCK) cells as a model.

Methods: MDCK cells were cultured on collagen film in the trans-well chamber and FLAG-tagged MT1-MMP and its mutants were expressed, and their localization were analyzed by confocal microscopy. Tubulogenesis was studied by culturing MDCK cells in 3D collagen gel with stimulation of hepatocyte growth factor (HGF) and/or transforming growth factor beta (TGFbeta).

Results: Polarized MDCK cells cultured on fibrillar collagen exclusively localized MT1-MMP to the apical side of their plasma membrane. Upon treating cells with HGF, which stimulates tubulogenesis of MDCK cells in 3D collagen, MT1-MMP was localized to the basal side, resulting in efficient collagen degradation. In 3D collagen MT1-MMP localized to basal side of cells that were extending into the collagen gel, but cells in non-extending parts of the structure did not localize MT1-MMP to their basal side. TGFbeta is spontaneously expressed in MDCK cells and has been shown to inhibit tubulogenesis. We found that addition of TGFbeta at 10 ng/ml inhibited both tubulogenesis and basal localization of MT1-MMP, indicating TGFbeta is indeed a negative regulator. However, inhibition of TGFbeta signaling rather inhibited both tubulogenesis and MT1-MMP localization to the basal side. Interestingly, lower dose of TGFbeta at 100 pg/ml enhanced both tubulogenesis and basal localization of MT1-MMP, indicating that action of TGFbeta is biphasic.

Conclusion: Taken together, differential local TGFbeta signaling may be a key to regulate local MT1-MMP levels at the basal side of the epithelial cells, resulting in formation of tube structure.