

INTERMOLECULAR COMPLEXES OF MT1-MMP, Cdc42 AND $\alpha 2\beta 1$ EC LUMEN FORMATION IN 3D COLLAGEN MATRICES

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A major focus of our laboratory is to elucidate molecular mechanisms controlling the ability of endothelial cells (ECs) to form luminal structures during vascular morphogenesis in 3D collagen matrices. EC luminal and tube formation is particularly dependent on a signaling axis involving the collagen-binding integrin $\alpha 2\beta 1$, the Rho GTPases, Cdc42 and Rac1 and the membrane-type 1 metalloproteinase, MT1-MMP. This information was obtained using blocking antibody experiments directed to $\alpha 2\beta 1$, dominant negative mutants for Cdc42 and Rac1, siRNA suppression for all four molecules and proteinase inhibitors for MT1-MMP. In all four cases, blockade of function of these molecules inhibits EC lumen formation. How these molecules signal together to control vascular morphogenesis in collagen matrices is not well understood and is the subject of considerable investigation in our laboratory. To begin to address this question, we hypothesized that these proteins may exist in intermolecular complexes that might be regulated during EC lumen and tube formation. Using either epitope-tagged MT1-MMP or Cdc42 we were able to specifically capture each of the above endogenous proteins from ECs, suggesting that they work in conjunction to promote lumen formation. Furthermore, we see a stronger association of these intermolecular complexes during EC tube formation in 3D collagen matrices (compared to 2D matrices) which is in part controlled by EC interactions with collagen and other matrix components. These data suggest that intermolecular complexes of MT1-MMP, Cdc42 and integrins control EC lumen formation during vascular morphogenesis.