ABSTRACT

Granzyme B (GZMB) is a serine protease that is expressed by a variety of immune cells and is abundant in a large number of chronic inflammatory disorders. GZMB is highly expressed in cytotoxic lymphocytes where it serves as the main effector molecule of the granule exocytosis pathway by which cytotoxic immune cells mediate target cell death through intracellular delivery of GZMB, leading to activation of apoptotic signaling cascades. GZMB can also accumulate extracellularly during inflammation, where it can cleave a range of extracellular matrix (ECM) proteins that may disrupt cell-matrix interactions and modulate the bioavailability of matrix-bound growth factors. In this dissertation I have explored the intracellular and extracellular roles of GZMB in vascular remodeling in disease. By examining human atherosclerotic plaques, I discovered an imbalance between GZMB and its endogenous inhibitor, proteinase inhibitor 9 (PI-9). PI-9 expression by vascular smooth muscle cells (VSMC) in plaques was reduced with increased disease severity. Elevated levels of GZMB in advanced lesions were correlated with reduced PI-9 expression and increased VSMC apoptosis. These findings suggest that VSMC are more susceptible to GZMB-induced apoptosis in advanced lesions due to reduced PI-9 expression. While examining the extracellular activities of GZMB on vascular remodeling, I focused on the role of GZMB-mediated cleavage of fibronectin (FN), a known GZMB substrate. FN has a major role in regulating angiogenesis as it facilitates endothelial cell (EC) migration and capillary formation, as well as binding to angiogeninc growth factors in the ECM including vascular endothelial growth factor (VEGF). VEGF is a potent vascular permeabilizing agent that is sequestered in the ECM by binding FN. GZMB-mediated FN cleavage resulted in reduced EC adhesion, migration and capillary tube formation. In addition, GZMB-mediated FN cleavage induced the release of VEGF from the ECM and promoted VEGF-dependent vascular leakage *in vivo.* Thus, GZMB may contribute to the progression and/or persistence of chronic inflammation by dysregulating angiogenesis and promoting vascular permeability. Collectively, the results of this work suggest that both intracellular and extracellular GZMB activities contribute to vascular remodeling and pathological angiogenesis