E-selectin functions in venous thrombosis by binding and activating host cells to initiate the coagulation cascade. The E-selectin antagonist, Uproleselan (GMI-1757), has been shown to attenuate thrombus formation following electrical stimulation in a preclinical inferior vena cava (IVC) model without significantly affecting hemostasis (Culmer DL et al. Thromb Haemost. 117:1171-1181, 2017). A recent study has reported that galectin-3 (gal-3) and gal-3 binding protein are associated with murine thrombogenesis where they interact at the thrombus-vein wall interface, and that gal-3 may be contributing to thrombosis through proinflammatory mechanisms (Elise P et al. Blood 125:1813-1821, 2015). Collectively these studies suggest that pharmacologic targeting of both E-selectin and gal-3 function may afford a new class of therapeutics for treatment of venous thrombosis. In the present studies, we report on the activity of a novel glycomimetic compound with dual functional antagonism of E-selectin and gal-3, and to demonstrate its anti-thrombotic activity in an IVC model.

Results

E-selectin IC50 = 798.1 nM

GMI-1757 was assessed for its ability to inhibit binding of galectin-3 binding protein using an ELISA format. Samples of 0.5 μg/ml solution of galectin-3 (B, America) in PBS/0.1% FBS were incubated with various concentrations of GMI-1757 and the mixtures were added to the wells of a 96-well plate that were coated with 1 μg/ml galectin-3 binding protein (R&D Catalog #520-GAN). After a two hour incubation at room temperature the wells were washed and the amount of bound galectin-3 was determined with an anti-galactin-3 HRP-conjugated antibody (R&D, Catalog #65755). The IC50 of GMI-1757 for galectin-3/galactin-3 binding protein interaction was determined.

Galectin-3 IC50 = 112.4 nM

GMI-1757 was assessed by its ability to inhibit binding of gal-3 to gal-3 binding protein using an ELISA format. Samples of 0.5 μg/ml solution of gal-3 (B, America) in PBS/0.1% FBS were incubated with various concentrations of GMI-1757 and the mixtures were added to the wells of a 96-well plate that were coated with 1 μg/ml gal-3 binding protein (R&D catalog #220-GAN). After a two hour incubation at room temperature the wells were washed and the amount of bound gal-3 was determined with an anti-gal-3 HRP-conjugated antibody (R&D, Catalog #65755). The IC50 of GMI-1757 for galectin-3/galactin-3 binding protein interaction was determined.

Summary: In comparison to the non-CNV eye, intravitreal administration of GMI-1757 led to a complete inhibition of laser-induced fibrosis.

Conclusion

An innovative dual antagonist of E-selectin and galectin-3, GMI-1757, has been produced that demonstrates marked attenuation of thrombosis formation in a murine IVC model. GMI-1757 is also shown to inhibit the development of fibrosis in a rat CNV model. Mechanism-of-action studies continue to be pursued to fully understand the impact of E-selectin and galectin-3 inhibition in this model and other models where disease progression is dependent on both inflammation and fibrosis.

References


Figure 1. GMI-1757 Antagonizes E-selectin and Galectin-3 Interactions

Figure 2. GMI-1757 Inhibits Galectin-3 Binding to Galectin-3 Binding Protein

Figure 3. GMI-1757 Inhibits Thrombosis in a Mouse Inferior Vena Cava Model

Figure 4. GMI-1757 Inhibits Fibrosis in a Rat Model of Choroidal Neovascularization

A NOVEL GLYCOMIMETIC COMPOUND (GMI-1757) WITH DUAL FUNCTIONAL ANTAGONISM TO E-SELECTIN AND GALECTIN-3 DEMONSTRATES INHIBITION OF THROMBUS FORMATION IN AN INFERIOR VENA CAVA MODEL

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