Abstract # LB164

A Novel Glycomimetic Compound (GMI-1757) with Dual Functional Antagonism to E-selectin and Galectin-3 Attenuates Fibrosis, Facilitates Mononuclear Cell Infiltration and Optimizes anti-PD-L1 Therapeutic Activity in a Pancreatic Adenocarcinoma Model

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Abstract

The inability to generate robust anti-tumor responses with therapeutics affecting T cell function has been ascribed to multiple parameters such as the tumor fibrotic compartment and the degree/makeup of host infiltrative cells. These parameters are affected through pathways involving the interaction of galectin-3 and E-selectin with cognate ligands. In the current investigations we assessed the impact of a dual E-selectin and galectin-3 glycomimetic antagonist, GMI-1757, in combination with anti-PD-L1 on tumor progression using an orthotopic pancreatic carcinoma model. Eight days following the orthotopic injection of $5x10^5$ luciferase-enabled Pan02 carcinoma cells into the pancreas of female C57BL/6 mice, mice began daily IP treatment for 11 days with control diluent (C) or 50 mg/kg GMI-1757. Beginning on study day 15 C- or GMI-1757-treated mice were given 10 mg/kg anti-PD-L1 or an isotype control antibody (LTF-2). Sixteen mice were allocated into each of 4 groups: group 1 = C + LTF-2; group 2 = GMI-1757 + LTF-2; group 3 = C + anti-PD-L1; and group 4 GMI-1757 + PD-L1. Bioluminescence imaging (BLI) intensities were determined over the course of the study (Day 0 to 26) to assess anti-tumor activity, and tissue from was excised on study days 15 and 27 (study conclusion) to assess fibrosis and immune infiltration. Tumor growth progressed in mice treated with LTF-2 in combination with C or GMI-1757 (final %T/C of 100 and 60.7, respectively), and appeared arrested in mice treated with C and anti-PD-L1 antibody (%T/C = 18.3). In contrast, treatment of tumor-bearing mice with GMI-1757 in combination with anti-PD-L1 antibody resulted in tumor regressions in 5/8 mice with a final %T/C = 0.60. Using a histopathologic scoring system, the extent of fibrosis within the primary pancreatic tumors was dependent on treatment with GMI-1757. Tumor fibrosis was moderate to marked in groups 1 and 3, and minimal to mild in groups 2 and 4. These results were confirmed by morphometric analysis of Masson's Trichrome stained sections where the percent tumor fibrosis area was approximately 29 and 19% in groups 1 and 3, respectively, and 10 and 5% in groups 2 and 4, respectively. Notably, tumor infiltrating immune cells were enhanced with GMI-1757 treatment as compared to C (moderate to marked vs. minimal). The immune phenotypes comprising the increased mononuclear cell infiltration into orthotopic Pan02 tumors continues to be investigated. In summary, we report on the use of a dual E-selectin and galectin-3 antagonist to attenuate fibrosis and enhance mononuclear cell infiltration in an orthotopic model of pancreatic carcinoma. The shifts in fibrotic response and cellular infiltration obtained with GMI-1757 treatment in this model creates a more robust antitumor effect when combined with anti-PD-L1 treatment compared to anti-PD-L1 treatment alone. The impact of GMI-1757 to combine with immune modulators where fibrosis and restricted host cell infiltration negatively impact tumor response continues to be investigated.

Background

- The microenvironment of pancreatic ductal carcinoma is a dynamic network composed of a highly fibrotic interstitium containing tumor cells, extracellular matrix components, and immune-inflammatory cells
- The interaction of galectin-3 and E-selectin with cognate ligands are known to impact fibrotic development and host inflammatory cell infiltration preventing the generation of robust anti-tumor responses with therapeutics affecting T cell function
- GMI-1757 is a rationally designed glycomimetic with dual inhibitory specificity to E-selectin and galectin-3 (K_D = 396 and 86 nM, respectively as determined by microscale thermophoresis)
- An orthotopic pancreatic ductal carcinoma model was used to investigate the tumor response to a combination of GMI-1757 and anti-PD-L1 with a special reference to the development of fibrosis and host cell infiltration

GlycoMimetics, Inc.

Results

Figure 1. Individual Pan02 Tumor Growth Curves assessed by BLI Imaging of Treatment Groups

Beginning eight days post implantation of $5x10^5$ luciferase-enabled Pan02 into the tail of the pancreas of female C57BL/6 mice, animals were randomized into four treatment groups and received: A) vehicle control (IP, bid days 8-18) and 10 mg/kg isotype control antibody (IP, qd days 15,18, 22 and 25); B) 50 mg/kg GMI-1757 (IP, bid days 8-18) and 10 mg/kg isotype control antibody (IP, qd days 15,18, 22 and 25); C) vehicle control (IP, bid days 8-18) and 10 mg/kg anti-PD-L1 antibody (IP, qd days 15,18, 22 and 25); and D) 50 mg/kg GMI-1757 (IP, bid days 8-18) and 10 mg/kg anti-PD-L1 antibody (IP, qd days 15,18, 22 and 25). BLI was conducted on study days 8, 15, 22 and 26.

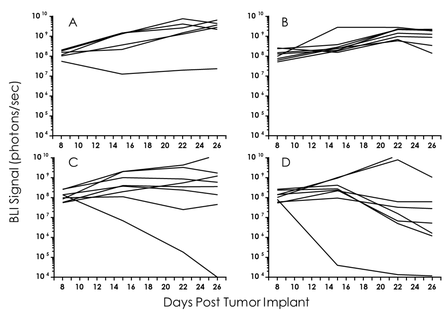
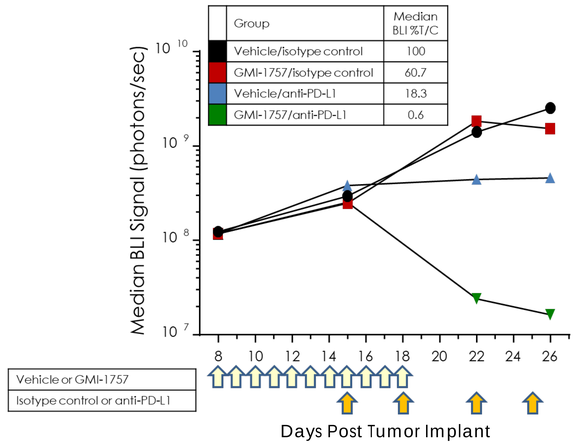


Figure 2. GMI-1757 Administration Augments the Response of Orthotopic Pan02 Pancreatic Tumors to Treatment with anti-PD-L1



Summary (Figures 1 and 2)

- Treatment with GMI-1757 in combination with anti-PD-L1 resulted in a 50% incidence of partial regressions and a 12.5% incidence of complete regression, with an approximate 99% reduction in median tumor volume as measure by BLI at study termination
- In the absence of GMI-1757 administration, treatment with anti-PD-L1 resulted in a 25% incidence of partial regressions, a 12.5% incidence of complete regression, with an approximate 80% reduction in median tumor burden

Results (cont.)

Figure 3. Treatment of Tumor Bearing Mice with GMI-1757 or GMI-1757 and anti-PD-L1 Decreased the Incidence of Intratumoral Fibrotic Development and Increased Mononuclear Infiltration

On study day 27, all mice were euthanized, pancreatic tumor was excised, bisected and fixed in 10% NBF. Tumors were processed, embedded in paraffin blocks, sectioned and stained with hematoxylin and eosin or Masson's Trichrome. The slides were blinded and evaluated for the microscopic incidence of fibrosis, necrosis, apoptosis, neutrophil infiltration and mononuclear infiltration.

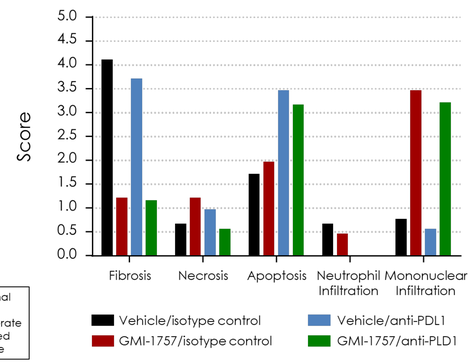
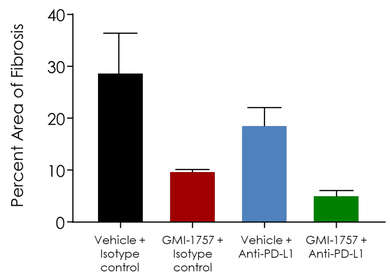


Figure 4. Treatment of Extent of Fibrosis Following GMI-1757 Treatment in Combination with Anti-PD-L1

Percent area of fibrosis in tumors was quantified by image analysis on Masson's trichrome stained sections of pancreas. Using Visopharm software, total tumor area and area of fibrosis were measured and the percent area of fibrosis was calculated (area of fibrosis/total tumor area x 100).



Conclusions

- Treatment with GMI-1757, an E-selectin/galectin-3 antagonist, in the orthotopic Pan02 tumor model resulted in a decrease in fibrosis and augmented immune response, as evidenced by an increase in intratumoral mononuclear cell infiltration
- Addition of GMI-1757 to anti-PD-L1 treatment resulted in enhanced anti-tumor effects, as compared to anti-PD-L1 treatment alone
- The impact of GMI-1757 with immune modulators where fibrosis and/or restricted immune cell infiltration negatively impact tumor response continues to be investigated