Background: Cancer immunotherapies, including checkpoint blockade antibodies, represent a breakthrough in cancer treatment with long-term remission observed in select patients. Unfortunately, many tumors remain unresponsive to existing immunotherapies, creating an urgent need to therapeutically target additional immune checkpoints that drive cancer progression. Glyco-immune checkpoints – in which cell-surface biopolymers decorated with sugars, or glycans, engage glycan-binding receptors on immune cells – have emerged as immune modulatory pathways that are misregulated in the context of cancer. In particular, recent evidence suggests that upregulation of the sialic acid monosaccharide on cancer cell surfaces allows tumors to resist treatment by engaging inhibitory receptors called Siglecs on immune cells. However, the lack of glycan-binding reagents with high affinity and selectivity has historically prevented targeting of tumor-associated glycans for cancer immunotherapy.

Methods: To address this need, we have developed a new class of antibody-lectin bispecifics (AbLecs) targeting tumor-associated glycans for checkpoint blockade (figure 1). In this approach, glycan-binding domains from immune receptors (e.g., Siglec receptors) are coupled to high-affinity binding domains from FDA-approved antibodies targeting common tumor-associated antigens (e.g., trastuzumab, rituximab, cetuximab) via knobs-into-holes heterodimerization technology.

Results: Western blot and mass spectrometry results indicated correct heterodimeric assembly of eight AbLec candidates, demonstrating the feasibility of the AbLec approach. We found that AbLecs bind cancer cell lines at nanomolar concentrations and block cognate Siglec receptor binding via flow cytometry. Finally, we showed that AbLecs enhance antibody-dependent phagocytosis and cytotoxicity of diverse human tumor cell lines by primary immune cells in vitro compared to their FDA-approved parent antibodies. Enhancement of in vitro tumor cell killing with AbLecs was dependent on the targeted Siglec and was more potent than the combination of the parent monoclonal antibody and a Siglec-blocking antibody.

Conclusions: These studies provide proof-of-principle for AbLecs as a first-in-class, modular platform technology enabling blockade of glyco-immune checkpoints for cancer immunotherapy.