INV322, A TME SELECTIVE CD25 X CTLA4 BISPECIFIC ANTIBODY APPROACH FOR DEPLETION OF TUMOR RESTRICTED TREGS

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Background Tregs maintain immune homeostasis in healthy individuals by limiting excessive or aberrant immune responses to environmental or self-triggers. In the context of tumor immunity, Tregs are associated with increased tumor progression, poor patient prognosis, and limited responsiveness to immunotherapeutic approaches. Targeting of Tregs has shown promise in the clinic, though current approaches are limited by on-target, off-tumor induction of autoimmune related toxicities associated with global Treg blockade. To overcome these toxicities and improve efficacy, Invenra has generated INV322, a bispecific antibody designed to preferentially engage tumor microenvironment (TME) Tregs. By targeting Treg co-expressed targets, CD25 and CTLA-4 with lower-affinity monovalent interactions, INV322 is designed to drive avidity only in the presence of dual target engagement to support selective blockade of Treg function and depletion by Fc-mediated clearance.

Methods INV322 is a human-targeted bispecific antibody, engineered using Invenra’s fully human B-Body® platform with a wild-type IgG1 for Fc-gamma-mediated effector function. Specificity of INV322 to CD25 and CTLA-4 was evaluated using both solid phase and cell-based read-outs. Potency of INV322 was evaluated by flow cytometry in vitro on overexpression cell lines and iTregs via NK-mediated cytotoxicity assays. A surrogate molecule, INV323, with specificity to murine CTLA-4 and CD25 was generated to evaluate in vivo potency in murine solid tumor models.

Results INV322 demonstrated selective lower-affinity monovalent binding to cells expressing CTLA-4 (single-digit-nM) and CD25 (triple-digit nM) independently. However, when bound to cells expressing both targets, these monovalent affinities result in combined avidity and result in improved sub-nM binding of INV322 to these cells. The strength of the avid binding was associated with improved Fc-mediated depletion as the potency of INV322 was increased on engineered lines expressing both targets compared with cell lines expressing single targets. In vivo evaluation of INV323 demonstrated Fc-dependent anti-tumor activity, and additive effects in combination with anti-PD-1 treatment after single – dose administration. The tumor-protective activity observed was associated with establishment of anti-tumor memory and correlated to Treg depletion and an increase in the Teff:Treg ratio within the TME.

Conclusions INV322 is an innovative bispecific approach for the selective depletion of tumor resident Tregs. The unique binding profile is designed to direct selective engagement of Tregs within the TME, providing a novel avenue for potential improvement of therapeutic index and patient outcome as compared to current Treg modulating therapeutics. IND-enabling activities are underway with INV322.

Ethics Approval All animal studies were reviewed and approved under Invenra’s IACUC protocol (I-RP-A-07).