There is no clear need for targeted therapies to treat acute myeloid leukemia (AML), the most common acute leukemia in adults. CD123 (IL-3 receptor alpha chain) is an attractive target for AML treatment [1]. However, cytotoxic antibody targeting CD123 proved to be insufficiently effective in a combination setting in phase III clinical trials [2]. T-cell engagers targeting CD123 displayed some clinical efficacy but were often associated with cytokine release syndrome and neurotoxicity [3]. Interest in the use of NK cells for therapeutic interventions has increased in recent years, as a potential safer alternative to T cells. Several NK-cell activating receptors can be targeted to induce antitumor immunity. We previously reported the development of trifunctional NK cell engagers (NKCEs) targeting a tumor antigen on cancer cells and co-engaging NKp46 and CD16a on NK cells [4]. We report here the design, characterization and preclinical development of a novel trifunctional NK cell engager (NKCE) targeting CD123 on AML cells and co-engaging NKp46 and CD16a on NK cells. We compared CD123-NKCE and a cytotoxic ADCC-enhanced antibody targeting CD123, in terms of antitumor activity in vitro, ex vivo and in vivo. Pharmacokinetic, pharmacodynamic and safety profiles of CD123-NKCE were evaluated in non-human primates (NHPs).

1. CD123-NKCE: strong cytotoxic activity against AML cells independently of high-affinity FcRn expression on AML cells

The expression of high-affinity FcRn CD64 on AML cells inhibits the ADCC activity of ADCC-enhanced antibody (CD123-lgG1+) in vitro (A) and ex vivo (B) but does not affect CD123-NKCE killing activity.

2. CD123-NKCE: strong potency against MOLM-13 AML cells in vitro

Co-engagement of NKp46 + CD16 for optimal NK cell activation

Cytotoxicity of NK cells against MOLM-13 AML cells

3. CD123-NKCE promotes NK cell activation, effector cytokine / chemokine production in vitro

CD123-NKCE promotes NK cell activation (CD57a, CD90) and effector cytokines (TNFa & IFNp) / chemokine (MIP3) production in vitro only in presence of MOLM-13 target cells expressing CD123.

4. CD123-NKCE: potent anti-tumor activity in mice model superior to anti-CD123 ADCC-enhanced IgG1 and NK cell-dependent

Deposition of NK cells in vivo abrogates CD123-NKCE mediated anti-tumor effect

5. CD123-NKCE leads to minimal cytokine release in vitro in human PBMC as compared to T-cell engager

6. CD123-NKCE: Pharmacokinetics (PK) exposure and Pharmacodynamics (PD) effect in NHP is associated with very limited cytokine release and no clinical signs

Complete depletion of CD123+ immune cells in NHP at low & high doses

Target Mediated Drug Disposition (TMDD) observed at low-dose

PD effects are associated to minimal pro-inflammatory cytokine release (<50 pg/ml).

Conclusion

- The expression of the high affinity Fcg receptor CD64 on patient-derived AML cells inhibited the ADCC activity of the antibody targeting CD123 in vitro and ex vivo, but not the antitumor activity of CD123-NKCE.
- CD123-NKCE led to potent antitumor activity against primary AML blasts and AML cell lines, promoted strong NK-cell activation and induced cytokine secretion only in the presence of AML target cells.
- Its antitumor activity in a mouse model was greater than that of the comparator antibody and dependent on the presence of NK cells.
- Moreover, CD123-NKCE generated strong and prolonged pharmacodynamic effects in NHP at very low doses, was well-tolerated up to 3 mg/kg and triggered only minor cytokine release.
- The data for activity, safety, pharmacokinetics, and pharmacodynamics provided here demonstrate the superiority of CD123-NKCE over the comparator cytotoxic antibody, in terms of antitumor activity in vitro, ex vivo, and in vivo, and its favorable safety profile, as compared to T-cell therapies.
- These results demonstrate the efficacy of CD123-NKCE for controlling AML tumors in vivo, and provide consistent support for the clinical development of IPH6101 / SAR443576.