LIF as a novel cancer immunotherapy target: modulating the tumor microenvironment with MSC-1, a humanized anti-LIF monoclonal antibody

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Abstract

Leukemia Inhibitory Factor (LIF) is a pleiotropic cytokine involved in many physiological and pathological processes. LIF is highly expressed in a subset of tumors across multiple tumor types and has been shown to correlate with poor prognosis. LIF is hypothesized to contribute to tumor growth and progression by acting on multiple aspects of cancer biology, including immunosuppression of the tumor microenvironment and is a key regulator of cancer initiating cells (CICs). MSC-1, a first-in-class, humanized monoclonal antibody (IgG1), is a potent and selective inhibitor of LIF. MSC-1 leads to STAT3 inhibition by disrupting LIF signaling through the LIF receptor (LIFR). Blocking LIF with MSC-1 decreased tumor growth in multiple mouse tumor models and drove reprogramming of the tumor microenvironment through modulation of immunosuppressive macrophages and of several immune cell types. These findings form the basis of a robust therapeutic hypothesis, whereby MSC-1 treatment may lead to clinical activity in multiple cancer indications. Clinical testing is planned to initiate in the end of 2017 and trials will incorporate target engagement and PD biomarkers as well as efficacy endpoints

Background

LIF plays a central role in self renewal and immunosuppression

Hijacking a developmental program

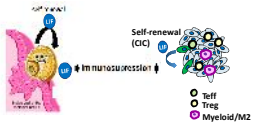

Figure 1A. Hypothesized mechanism of LIF activity in cancer

LIF: highly overexpressed and correlates with poor prognosis across multiple tumor types

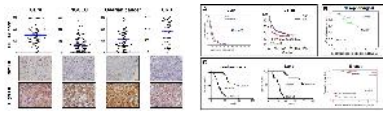

Figure 1B. IHC with an anti-LIF polyclonal antibody evaluating different tumor types (Blue bars represent mean +/- SEM). **Figure 1C.** A) LIF transcript levels in GBM (Seoane unpublished) and colon cancer [Yue 2015). B) Serum LIF protein levels in nasopharyngeal cancer [Liu 2013]. C) Intratumoral LIF protein in head and neck, lung and breast cancer [Albrengues 2014, Li 2014].

Results

MSC-1 is a potent function blocking LIF antagonist

- Humanized (from rat) function blocking antibody, IgG1 subclass
- Binds to LIF with high affinity (KD 64 pM by BiaCore) and specificity
- Does not bind to the most highly related IL-6 family member (OSM) or to other family members (CT-1, CNTF, CLC) capable of binding to the LIFR/gp130 heterodimer
- Inhibition of pSTAT3 signaling in U251 GBM and HCC1954 breast tumor lines in vitro (IC50 < 4 nM)
- In vivo efficacy in multiple mouse tumor models (GBM, lung, colon, ovarian) with parental rat and humanized MSC-1

MSC-1 molecular mechanism of inhibition

- LIF is a member of the gp130 family of cytokines
- LIF binding to LIFR/gp130 complex leads to:
  - -phosphorylation of Stat3 by JAK kinases
  - -pStat3 translocation to the nucleus
  - -transcription of Stat3 responsive genes

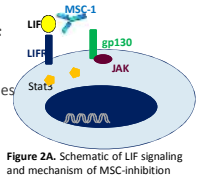

Figure 2A. Schematic of LIF signaling and mechanism of MSC-inhibition

Figure 2B. MSC-1 blocked phosphorylation of pSTAT3 in in U251 cells endogenously expressing LIF

Molecular MOA

MSC-1 molecular mechanism of inhibition

- **MSC-1 – function blocking anti-LIF antibody**
  - binds to LIF
  - blocks recruitment of gp130, Stat3 phosphorylation and downstream signaling
  - does not block binding of LIF to LIFR chain

Overlay of LIF complex structures

MSC-1 epitope detail

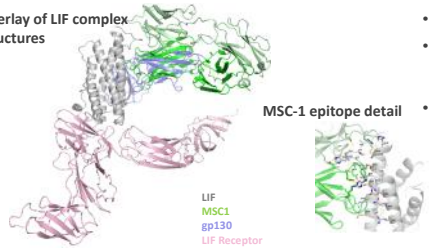

Figure 3. Crystal structure (solved to 3.1 Å) of MSC-1 Fab binding to LIF highlights specific interactions between antibody and LIF. The LIF binding site is overlapping with the previously identified gp130 binding site (Boulanger et al (2003) Mol.Cell 12: 577-589)

In Vivo Results

MSC-1 inhibits tumor growth in multiple syngeneic models and reprograms the TME

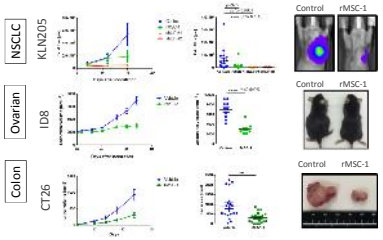

Figure 4. . Tumor growth in rMSC-1 and control treated mice in three syngeneic models, KLN205 lung tumors grown orthotopically, ID8 ovarian tumor (peritoneum) and CT26 colon cancer s.c. Vehicle control or rMSC-1 was administered IP 300 µg (~15 mg/kg) twice weekly

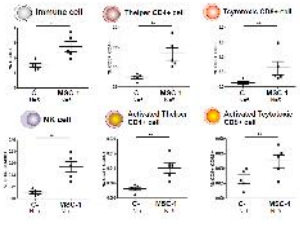

Figure 5. Immune cell infiltrates in ID8 tumors from rMSC-1 or control treated mice. Tumors (n=5) were harvested at d40 post implantation and were analyzed by flow cytometry. Similar results were observed in the KL205 and CT26 models

Cellular MOA and translation

LIF drives differentiation of immunosuppressive macrophages Translation from mouse models to human cellular mechanism

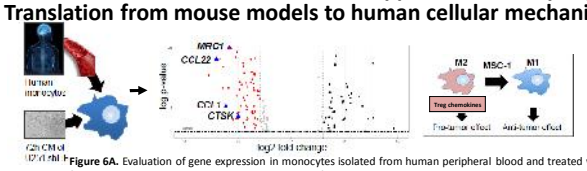

Figure 6A. Evaluation of gene expression in monocytes isolated from human peripheral blood and treated with conditioned medi from U251 cells and U251 cells with a knockdown of LIF expression by shRNA. The expression analysis is shown as a volcano plot highlighting differentially expressed M2 macrophage genes.

GBM patient organotypic slice – MSC1 treated

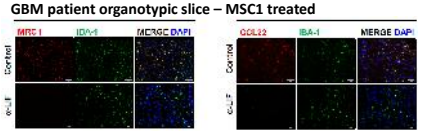

Figure 6B. Human GBM organotypic slices were incubated with rMSC-1 for 72 hrs. and stained using double immunofluorescence for (A) CD206 (MRC1) or (B) CCL22 with IBA-1. Representative images of the immunofluorescence are shown (scale bar, 50 µm). Similar results were observed using three patients

Conclusions

MSC-1 Proposed Mechanism of Action: a *pleiotropic MOA in cancer*

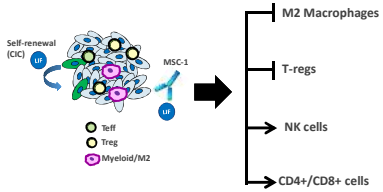

Figure 7. Schematic highlighting mechanism of action of LIF inhibition. Data from pre-clinical studies support a dual role for MSC-1 in blocking cancer initiating cell propagation and blocking immunosuppression in the tumor

LIF: a novel target and promising I/O approach with MSC-1 inhibition

- LIF is an IL-6 family member with a dual role promoting CIC renewal and immunomodulatory function
- Inhibition of LIF by MSC-1 is a promising approach to cancer therapy
- Pre-clinical data and a robust biomarker strategy will enable early evidence of target coverage and PD
- IND enabling studies have demonstrated a very good PK profile and no safety issues observed in toxicology studies
- MSC-1 is planned to enter Ph 1 clinical trials in Dec 2017/Jan 2018 with a robust biomarker strategy and plans to test in monotherapy and combinations