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.

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

Cellular MOA and translation

LIF drives differentiation of immunosuppressive macrophages

Translation from mouse models to human cellular mechanism

Figure 6A. Evaluation of gene expression profiles in primary cancer tissue samples from human peripheral blood and treated with conditioned media from U87 cells and U251 cells with a knockdown of LIF expression by shRNA. The expression analysis is shown as a volcano plot highlighting differential expression of 12500 unigenes.

Figure 6B. Human GBM organotypic slices were incubated with MSC-1 for 72 hs, and stained using double immunofluorescence for (A) CD206 (MRC1) and (B) CD112 with B2-1. Representative images of the immunofluorescence are shown (scale bar, 10 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 cell propagation and blocking immunosuppression in the tumor microenvironment.

MSC-1 as 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 LIF/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

- MSC-1 is a member of the gp130 family of cytokines
- LIF binding to gp130/gp130 complex leads to
  - phosphorylation of Stats and p70S6K
  - transcription of Stat3 responsive genes

In Vivo Results

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

Figure 5. Immune cell infiltration in U87 tumors from MSC-1 or control treated mice. Tumors (n=5) were harvested at day 35 and implanted in nude mice and were analyzed by flow cytometry. Similar results were observed in the KLu25 and CT26 models.

Figure 6C. Schematic illustrating mechanism of action of LIF inhibition. Data from pre-clinical studies support a dual role for MSC-1 in blocking cancer cell propagation and blocking immunosuppression in the tumor microenvironment.

Figure 1C. Overlay of LIF complex structures

Figure 3. Crystal structure (solved to 1.3 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) MolCell 12: 577-589.

Figure 4. Tumor growth in MSC-1 and control treated mice in three syngeneic models, KLu25 and CT26 lung tumors grown orthotopically, KLu25 ovarian tumor fragments) and CT26 colon cancer n.c. Vehicle control or MSC-1 was administered IP 300 μg/300 μg/300 μg/300 μg/300 μg weekly.

Background

LIF plays a central role in self renewal and immunosuppression Hijacking a developmental program

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

Figure 16. Hypothetical mechanism of LIF activity in cancer

Figure 10. MSC with an anti-LIF polyclonal antibody evaluating different tumor types (blue bar graph mean ± SEM). Figure 11. Inhibition of pSTAT3 signaling in U251 GBM and HCC1954 breast tumor lines in vitro (IC50 < 4 nM). Figure 12. Reduced LIF protein levels in nasopharyngeal cancer [Liu 2013]. C) Western blot of LIF levels in U87 and U251 GBM cell lines.

Results

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MSC-1 treated

- Differentiation of myeloid/M2 macrophages
- Blockage of recruitment of gp130
- STAT3 inhibition

Figure 2A. Schematic of LIF signaling and mechanisms of MSC-1 inhibition

Figure 4B. Schematic of LIF signaling and mechanisms of MSC-1 inhibition

Figure 2B. MSC-2 blocked phosphorylation of Stat3 (pY705) in U87 cells independently expressing LIF.

Figure 2C. MSC-2 blocked phosphorylation of Stat3 (pY705) in U87 cells independently expressing LIF.

Figure 2D. MSC-2 blocked phosphorylation of Stat3 (pY705) in U87 cells independently expressing LIF.

Figure 2E. MSC-2 blocked phosphorylation of Stat3 (pY705) in U87 cells independently expressing LIF.

Figure 2F. MSC-2 blocked phosphorylation of Stat3 (pY705) in U87 cells independently expressing LIF.

Figure 2G. MSC-2 blocked phosphorylation of Stat3 (pY705) in U87 cells independently expressing LIF.

Figure 2H. MSC-2 blocked phosphorylation of Stat3 (pY705) in U87 cells independently expressing LIF.

Figure 2I. MSC-2 blocked phosphorylation of Stat3 (pY705) in U87 cells independently expressing LIF.

Figure 2J. MSC-2 blocked phosphorylation of Stat3 (pY705) in U87 cells independently expressing LIF.