Development of OR2805, an anti-CD163 antibody derived from an elite responder to checkpoint inhibitor therapy that relieves immunosuppression caused by M2c macrophages

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Abstract # 271

Background: OR2805 antibody was discovered using B cells derived from an elite responder to checkpoint inhibitor (CPI) therapy. It is a fully human IgG1 antibody that binds to CD163, an immune-suppressive receptor highly expressed on tumor associated macrophages (TAMs). High numbers of CD163-expressing TAMs generally predict an unfavorable prognosis in solid tumors. These CD163-expressing TAMs contribute to an immune-suppressive tumor microenvironment and inhibit an anti-tumor T-cell response by engaging immune checkpoints and secreting immune-suppressive cytokines. Relieving the immune suppression of CD163-expressing TAMs to improve anti-tumor T-cell responses is a rational therapeutic strategy as monotherapy and in combination with CPI therapy.

Methods: Cocultures of immunosuppressive primary human polarized M2c macrophages with autologous CD8$^+$ T cells or phytohemagglutinin (PHA)-T cell blasts (exhausted T cells) were used to interrogate OR2805-dependent immunomodulatory responses as single agent and in combination with pembrolizumab, an anti-PD1 antibody. The anti-tumor activity of OR2805 was evaluated in humanized mouse models. Safety and pharmacokinetics (PK) profile of OR2805 was evaluated in cynomolgus monkeys and human whole blood for cytokine release assessment.

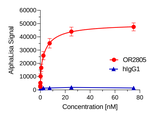
Results: In coculture assays, OR2805-treatment relieved the suppressive effect of M2c macrophages as demonstrated by increased T-cell proliferation and the release of IFN-γ and perforin. OR2805 restored the IFN-γ production of exhausted T cells and showed a synergistic effect on cocultures treated in combination with pembrolizumab. OR2805-treatment demonstrated significant anti-tumor activity in lung cancer xenograft models in humanized NSG-SGM3 mice. In cynomolgus monkeys, OR2805 demonstrated a typical IgG1 PK profile and good serum exposure. Furthermore, OR2805 did not trigger the release of IL-1β, IL-2, IL-4, IL-6, IL-10, IFN-γ, or TNF-α cytokines in whole blood from either healthy donors or NSCLC patients.

Conclusions: OR2805 reduced M2c-mediated immunosuppression and enhanced T cell effector functions. OR2805-treatment resulted in significant anti-tumor activity in lung cancer xenograft models in humanized mice. The pharmacology, PK, and toxicokinetic data support further development of OR2805 as an anti-cancer therapy, both as a monotherapy and in combination with CPI therapy.

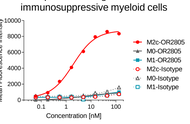
1

OR2805 demonstrates specific binding to immunosuppressive myeloid cells

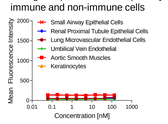
OR2805 binds to human CD163



Specific binding to human immunosuppressive myeloid cells



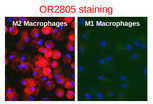
No binding to a panel of human primary immune and non-immune cells

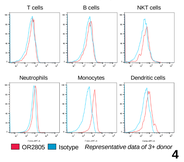


OR2805 binds to TAMs in dissociated NSCLC tumors

Cell surface markers	Patient 1 cells (%)	Patient 2 cells (%)
Total CD14$^+$ (monocytes)	26	30
CD163$^+$ of CD14$^+$ (M2c)	69	88
OR2805$^+$ of M2c	82	77
CD163$^-$ CD80$^+$ of CD14$^+$	20	11
OR2805$^+$ of CD163$^-$ TAMs	11	9

OR2805 staining





Representative data of 3+ donor

OR2805 has a potential to target immunosuppressive myeloid cells in the TME without impacting other cells

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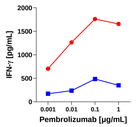
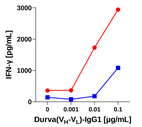
Combination with OR2805 enhances anti-PD-1 and anti-PD-L1 antibody activity in M2c/T cell blast coculture assays

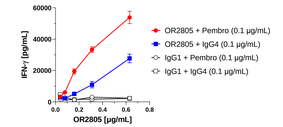
OR2805 + anti-PD-1



OR2805 + anti-PD-L1



Anti-PD-1 + OR2805



OR2805 (0.16 µg/mL) hIgG1 (0.16 µg/mL)

IFN-γ production is enhanced by combination of OR2805 with anti-PD-1 or anti-PD-L1 antibody in M2c/T cell blast coculture assays

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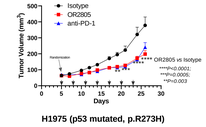
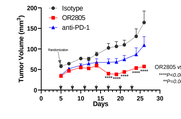
OR2805 induces robust anti-tumor activity in lung cancer xenograft models in humanized NSG-SGM3 mice

Tumor Volume

A549 (p53 WT)

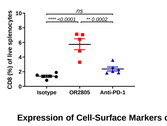


H1975 (p53 mutated, p.R273H)

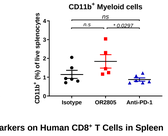


Proportions of Human T and Myeloid Cells in Spleen

CD8$^+$ T cells

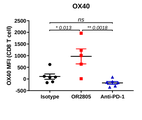


CD11b$^+$ Myeloid cells

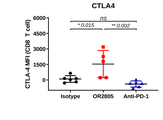


Expression of Cell-Surface Markers on Human CD8$^+$ T Cells in Spleen

OX40



CTLA4



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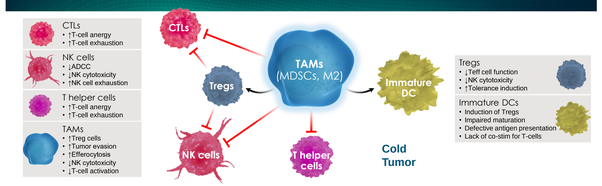
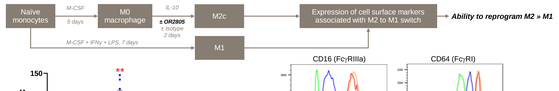
OR2805 targets TAMs in the TME to broaden and deepen responses

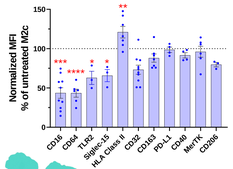


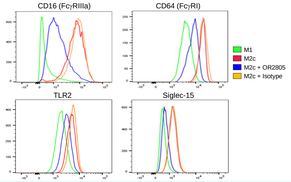
- M2 TAMs contribute to tumor immune evasion by suppressing anti-tumor immune responses and by promoting a tumorigenic milieu. Repolarization of suppressive myeloid cells to proinflammatory phenotype is an attractive strategy to enhance clinical responses to CPI therapy.
- OR2805 targets CD163 on immunosuppressive M2-like TAMs and relieves their suppressive effect leading to increased T cell activation and proliferation, T cell skewing towards Th1 phenotype, and enhanced T cell mediated killing of cancer cells. This reprogramming of TAMs may therefore enhance clinical responses to immunotherapy.

2

OR2805 reduces expression of M2c macrophage surface markers





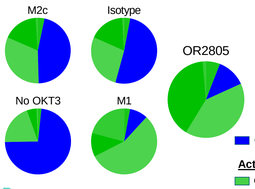


OR2805 treatment reduces expression of cell-surface markers associated with tumor-promoting M2c macrophages

5

OR2805-treated M2c macrophages skew T cells towards activated Th1-like phenotype

Distribution of CD4$^+$ T cells phenotypes



- CXCR3 is preferentially expressed on Th1 cells
- IFNγ production within the TME enhances the CXCR3-mediated T-cell recruitment to the tumor site
- CXCR3 signaling promotes CD8$^+$ T-cell infiltration
- CXCR3 expressing CD8$^+$ T-cell populations display enhanced cytotoxicity against tumor cells

Resting T cells
CXCR3$^+$ CD69$^-$ CD25$^-$

Activated CXCR3$^+$ T cells
CXCR3$^+$ CD69$^+$ CD25$^+$

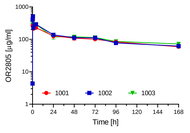
Activated CXCR3$^-$ T cells
CXCR3$^-$ CD69$^+$ CD25$^+$

OR2805-treated macrophages promote T-cell activation leading to greater expression of T-cell activation markers (CD69, ICOS, OX40)

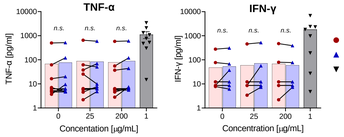
8

OR2805 toxicology predicts tolerable safety profile

PK in Cynomolgus Monkeys



Cytokine Release in Human Whole Blood



- Completed pilot, dose-range finding and GLP tox studies up to 100 mg/kg
- Observed OR2805 half-life in cynomolgus monkeys is about 5.8 days
- No in-life toxicity observed
- No abnormalities on pathological exam, and normal serum chemistries and hematology
- No changes in immune cell subsets
- Slight elevation in serum IL-6 suggesting biological activity
- OR2805 does not trigger cytokine release in whole blood from healthy human subjects

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CD163 - Normal physiology and role in cancer

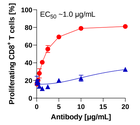
- Expression predominantly limited to immunosuppressive macrophages[1]
- Hemoglobin scavenger receptor upregulated on immunosuppressive macrophages
- Binding by its ligands induces secretion of immunosuppressive cytokines[2,3]
- Inhibits T-cell proliferation[4,5]
- Overexpression in human macrophages results in an M2 phenotype[6]
- Knockout mice develop normally however, lack protumoral activation of macrophages and fail to implant tumors[7]
- Expression in tumors correlates with poor survival[8-11]
  - In HNSCC, BC and GC, expression of CD163 correlated with decreased response to chemotherapy
  - Higher levels of expression in melanoma predicted poor response to CPI
  - CD163 expression correlates with IL-10 expression in melanoma

[1]Genomics Institute of the Novartis Research Foundation, [2]Molecular Immunology 2010;47:1650, [3]JCI Insight. 2016;1:e85375, [4]Biochem Biophys Res Commun. 2001;288:841,[5]Scientific Reports 2017;7:12940, [6]Immunobiology 2017;222:900, [7]Cancer Res 2018;78:3255, [8]Clin Transl Immunology 2020;9:e1108, [9]Cancer Management and Research 2020;12:5831, [10]Cell 2016;165:35, [11]J Exp Med. 2019;216:2394.

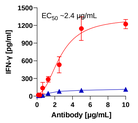
3

OR2805 treated M2c macrophages promote T-cell activation and proliferation

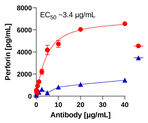
Proliferation



IFN-γ



Perforin



Representative data from n=11 donors

OR2805-treatment reduces the ability of M2c to suppress T-cell activation leading to greater T-cell stimulation (IL-2, IFNγ, TNFα, & perforin production), and both CD4$^+$ and CD8$^+$ T cell proliferation in coculture assays

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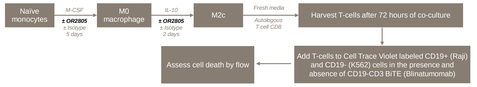
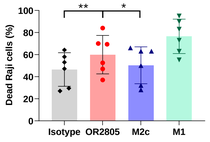
OR2805 cocultured CD8$^+$ T cells show enhanced ability to kill cancer cells

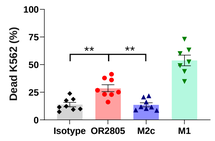


Raji B cell killing activity of CD8$^+$ T cells in the presence of CD19-CD3 BiTE



Composite data from n=6 donors

K562 cell killing activity of non-HLA restricted CD8$^+$ T cells



Composite data from n=8 donors

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Summary: OR2805 relieves immunosuppression caused by myeloid cells in the tumor microenvironment

- Binds with high specificity to M2 macrophages and TAMs in human primary NSCLC tumors
- Reduces expression of cell-surface markers associated with tumor-promoting M2c macrophages
- Minimizes M2 suppressive effect on T-cell activation and proliferation and skews T cells towards anti-tumor Th1 phenotype
- Shows enhanced expression of activation markers and cancer-killing ability in cocultured T cells
- Demonstrates robust anti-tumor activity in lung cancer xenograft models in humanized NSG-SGM3 mice
- Reduces TAM mediated immunosuppression and enhances anti-tumor immune responses
- Combination with OR2805 amplifies anti-PD-1 and anti-PD-L1 activity in coculture assays
- OR2805 toxicology predicts tolerable safety profile
- Phase 1/2 clinical study OR2805-101 open for enrollment

OR2805 reduces TAM-mediated immunosuppression and enhances anti-tumor immune responses, and has the potential as a single agent or in combination with CPI to increase the number of patients who may benefit from immunotherapy

THE UNIVERSITY OF TEXAS MD Anderson Cancer Center

Acknowledgements
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