Selective SIRPα blockade potentiates human dendritic cell antigen cross-presentation and triggers memory T cell anti-tumor responses

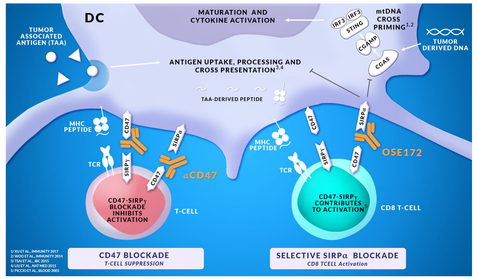
Vanessa Gauttier[1,2,3], Sabrina Pengam[1,2,3], Justine Durand[2,3], Aurore Morello[1,2,3], Virginie Vignard[4], Nathalie Labarrière[4], Sophie Conchon[2,3], Bernard Vanhove[1,2,3], Nicolas Poirier[1,2,3]

[1]OSE Immunotherapeutics, Nantes, France; [2]CRTI - UMR1064, INSERM, Université de Nantes, Nantes, France; [3]Institut de Transplantation Urologie Néphrologie (ITUN), CHU Nantes, Nantes, France; [4]CRCINA - UMR1232, INSERM, Université de Nantes, Nantes, France

AACR 2018
Poster #1684

Introduction

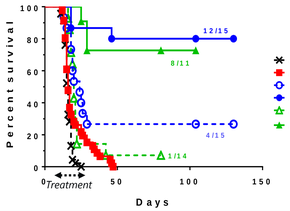
Interaction of SIRPα, expressed by myeloid cells, with the ubiquitous receptor CD47 is an important immune checkpoint of the innate response, involved in the regulation of myeloid functions including macrophage phagocytosis and dendritic cell antigen presentation. While anti-CD47 mAbs or SIRPα-Fc drugs target tumor cells and boost macrophage phagocytosis, here we show that combination therapies including selective targeting of SIRPα are efficient in mouse HCC tumor model by inducing robust memory anti-tumor T-cell responses (see poster #1753 for monotherapy effect). We also describe the opposite effect between anti-CD47 mAbs and selective SIRPα blockade in human that was not predictable from mouse studies on dendritic cell biology and antigen cross-presentation.



1 Anti-SIRPα combination with PD-L1 blockade or 4-1BB activation

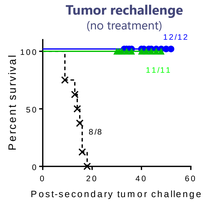
Induces high response rate and tumor elimination in orthotopic syngeneic HCC mouse model

Syngeneic hepatoma Hepa 1.6 cells were injected through the portal vein into C57Bl/6 male mice. Mice were treated three times a week for three weeks with a control monoclonal antibody (mAb) or with an antagonist anti-SIRPα mAb (P84) at 12 mg/kg i.p. The anti-PD-L1 mAb (10F-9G2) was injected two times a week (8mg/kg) for three weeks and the anti-4-1BB mAb (3H3) was injected twice at 4mg/kg (Day 4 & 8).
All treatments have been started four days after tumor inoculation.



2 Anti-SIRPα combinations induce anti-tumor memory responses

Durable and robust anti-tumor memory responses

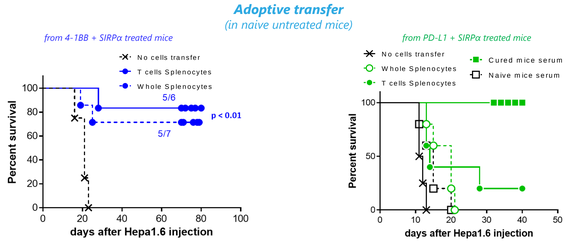


- Naïve mice
- cured mice SIRPa + PD-L1 mAbs
- cured mice SIRPa + 4-1BB mAbs

Mice previously cured with combination of anti-SIRPα + anti-PD-L1 or anti-4-1BB mAbs rejected spontaneously (without new treatment) a second intra-splenic tumor challenge.

Cellular and/or humoral anti-tumor memory responses

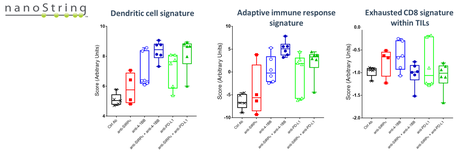
Adoptive transfer of splenocytes (10^7) or isolated T lymphocytes ($2.5x10^6$) from αSIRPα and α4-1BB Abs-treated and cured-mice protect naïve and untreated mice from intraportal injection of Hepa 1.6 cells. Surprisingly, from αSIRPα plus αPD-L1 Abs cured-mice, the transfer of serum (200µL) induced the elimination of the tumor in naïve mice whereas adoptive T-cell transfer demonstrated a very limited clinical effect.

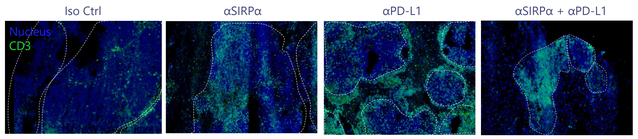


3 Anti-SIRPα modifies innate and adaptive tumor microenvironment

SIRPα blockade in combination increases non-exhausted T cells infiltration to the core of the tumor

HCC tumor bearing mice were treated as previously mentioned and euthanized thirteen days after tumor inoculation. Tumor microenvironment changes were evaluated by gene expression analysis and by immunostaining of CD3+ cells (green) on frozen tumor sections. Transcriptomic analysis of liver tumor was performed by NanoString and gene signatures were made by nSolver software.

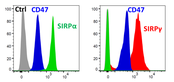


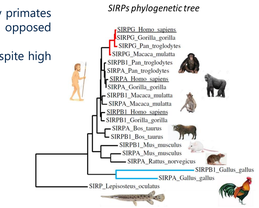


4 Selective SIRPα blockade enhances Ag presentation by dendritic cells

SIRPγ is absent in mice and is expressed by human T lymphocytes

SIRPγ, another CD47 ligand, is only expressed by primates and in a restricted manner on T lymphocytes as opposed to SIRPα expression on myeloid cells.
OSE-172 is a selective anti-SIRPα mAb which, despite high homology with SIRPα, does not bind to SIRPγ.

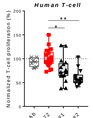


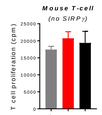


Anti-CD47 mAbs are immunosuppressive in humans

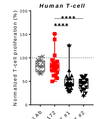
Anti-CD47 mAbs that block interaction with SIRPγ significantly inhibit human polyclonal or antigen-specific T-cell proliferation in human, not in mouse. In contrast, selective OSE-172 anti-human SIRPα mAb (does not bind SIRPγ) does not impede human T lymphocyte proliferation.

Polyclonal stimulation

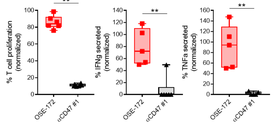




Allogeneic DC stimulation



Allogeneic PBMC stimulation

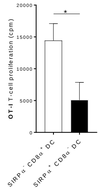


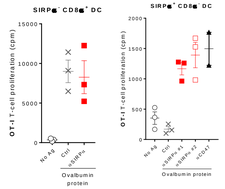
SIRPα and CD47 blockade enhance antigen cross-presentation **in mouse**

SIRPα+ DCs are less potent to cross-present antigen

Blocking at SIRPα or CD47 enhances Ag cross-presentation

SIRPα+ and SIRPα- DCs were isolated from spleen and cultured with OT-I CD8 T cells isolated from OVA specific TCR transgenic mice (OT-I), after loading with the ovalbumin protein overnight in presence of the different Abs.

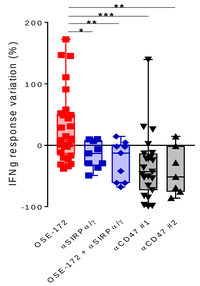




Only the selective blockade of SIRPα enhances tumor-associated antigen (TAA) cross-presentation **in human**

Method: HLA-A2+ monocytes were isolated from blood and differentiated into immature dendritic cells (DC) with GM-CSF and IL-4. After loading with a long peptide of the TAA Melan-A (25 mer) in the presence of the Abs, Melan-A loaded DCs were co-cultured with a CD8 T cell clone (isolated from Melanoma patients) specific of the HLA-A2/Melan-A complex. IFNγ intracellular staining was performed after a 5 hours-culture. Antibodies were added at 10 µg/ml and the IFNγ response was normalized to isotype control conditions.

Results: **While selective blockade of SIRPα with OSE-172 increased TAA cross-presentation by human DCs (as revealed by T-cell IFNγ response), anti-CD47 or non-selective anti-SIRPα/γ mAbs inhibited cross-priming.**



Conclusion

Selective blockade of SIRPα:

- Synergizes with checkpoint inhibitors or costimulatory agents
- Induces durable anti-tumor memory lymphocyte responses
- Modifies innate and adaptive tumor microenvironment

- Is not immunosuppressive on human T-cell responses and rather increases tumor-associated antigen cross-presentation by dendritic cells in humans.