Memorial Sloan Kettering Cancer Center

Targeting Tumor Metabolism to Overcome Resistance to Immune Checkpoint Blockade

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Immuno-Oncology Service
HOPP program, Depart of Medicine
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Prof. of Immunol. Research, Weill Cornell

sitc Society for Immunotherapy of Cancer

SITC 2022 Interim Workshop April 21 – 22, 2022 | San Diego, CA

LUDWIG CANCER RESEARCH

PARKER INSTITUTE FOR CANCER IMMUNOTHERAPY

SWIM ACROSS AMERICA
MAKING WAVES TO FIGHT CANCER

Disclosures

- IMVAQ therapeutics co-founder
- Advisory board immunos therapeutics, Algerian Research Agency in Health and Life Science
- Consulting for Pfizer, Daichii, Immunogensis, Kowa.
- Inventor on a patent applications related to work on Oncolytic Viral therapy, Alpha Virus Based Vaccine, Neo Antigen Modeling, CD40, GITR, OX40, PD-1 and CTLA-4.

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Kyn Therapeutics

Infinity Pharmaceuticals, Inc.

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Adaptive Biotechnologies

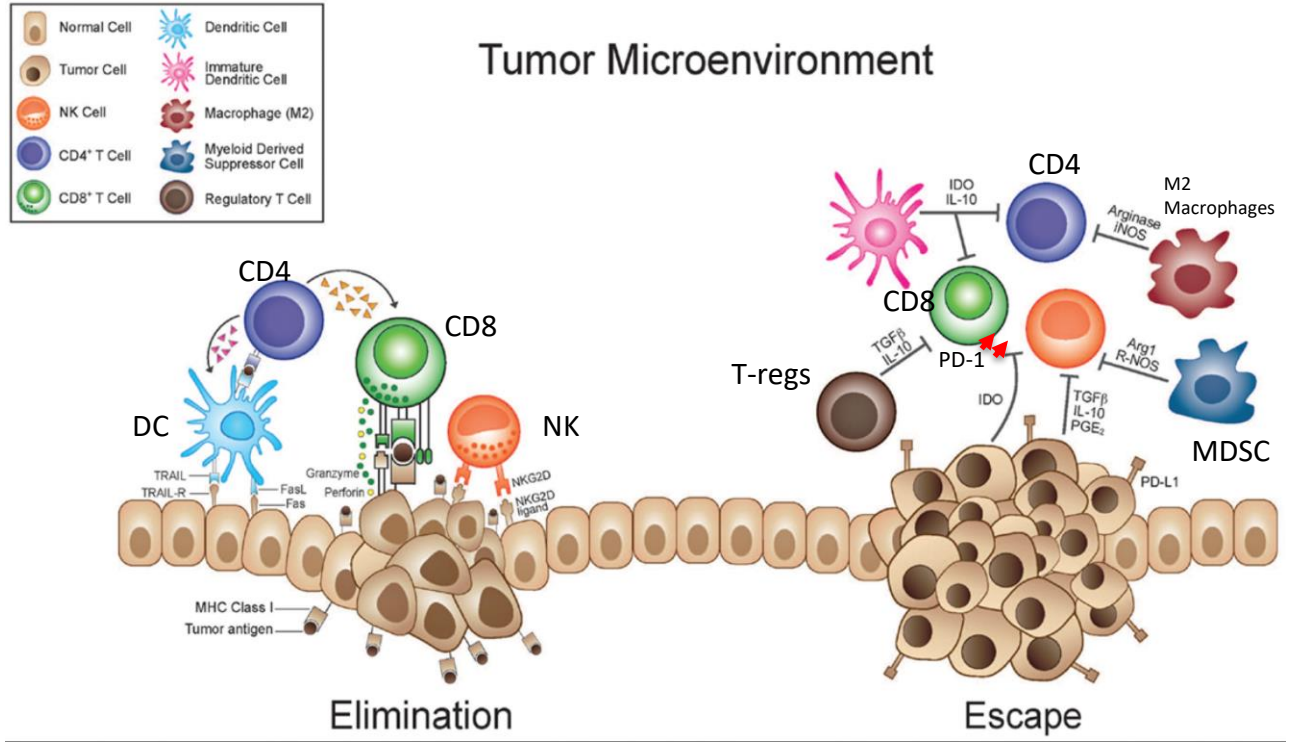
Leap Therapeutics, Inc.

Aprea.

Roche-Boehringer

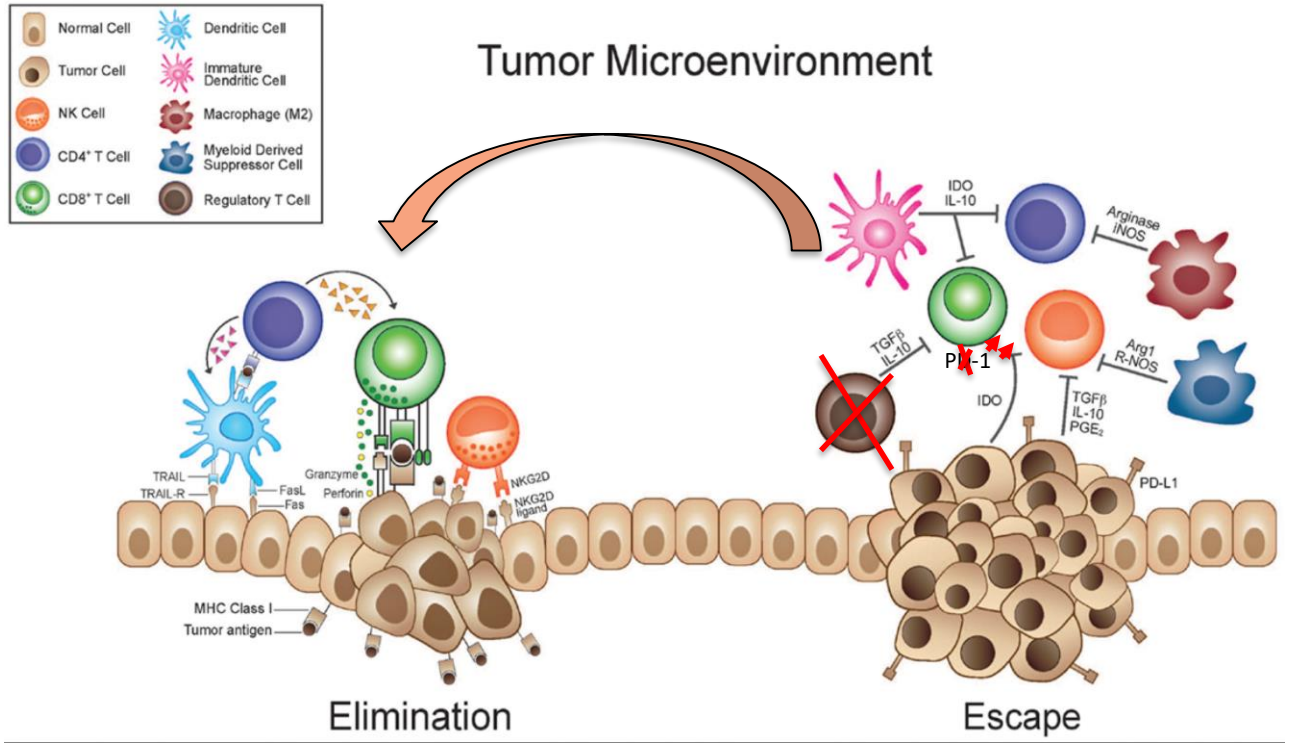
Immunoediting

Immune Suppressive Microenvironment



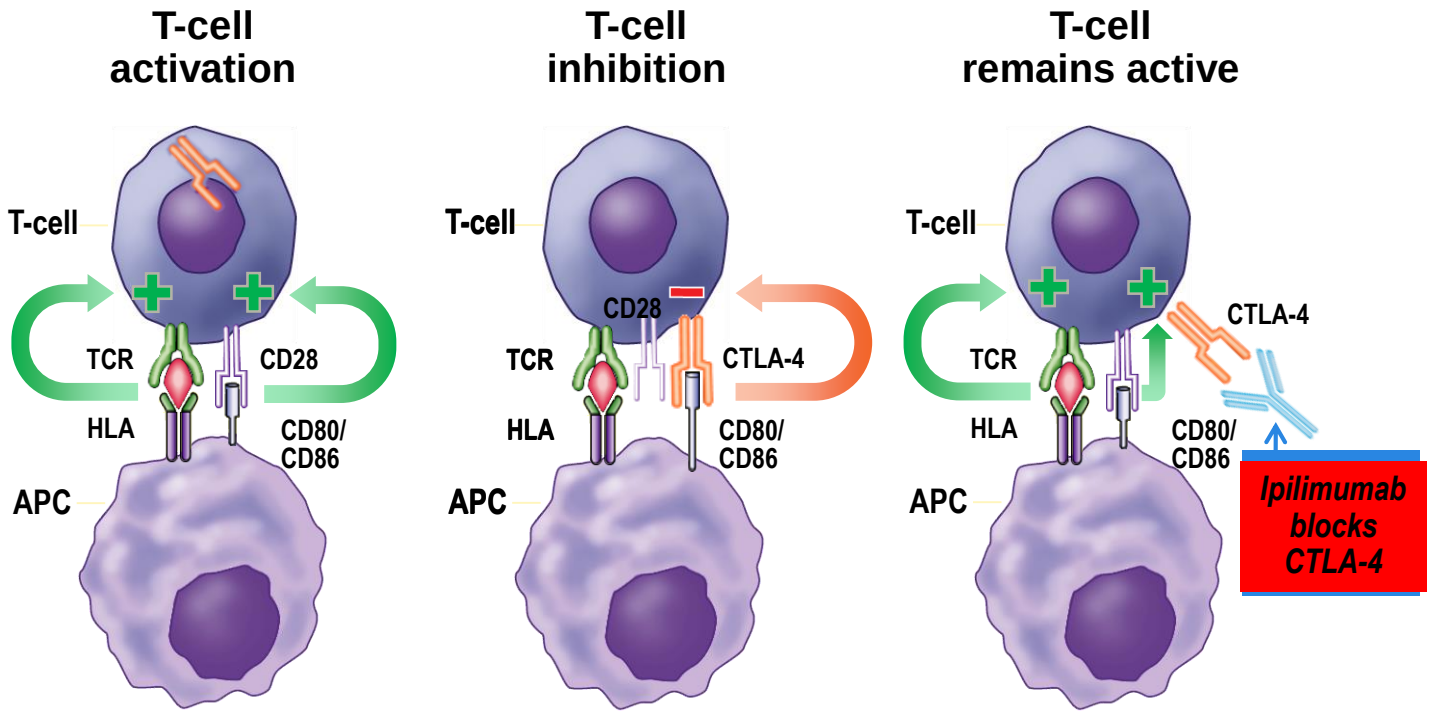
William J. Murphy. Front Oncol. 2013; 3: 197.

Immune Suppressive Microenvironment



William J. Murphy. Front Oncol. 2013; 3: 197.

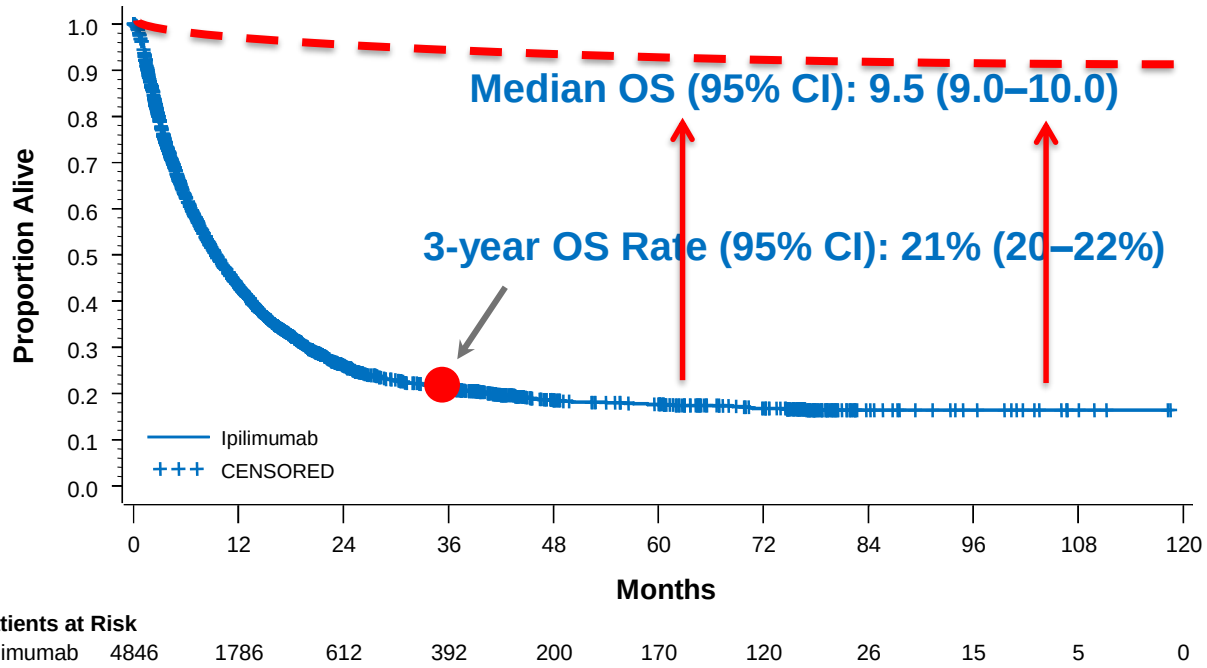
Ipilimumab Augments T-Cell Activation and Proliferation



Adapted from O'Day et al. Plenary session presentation, abstract #4, ASCO 2010.

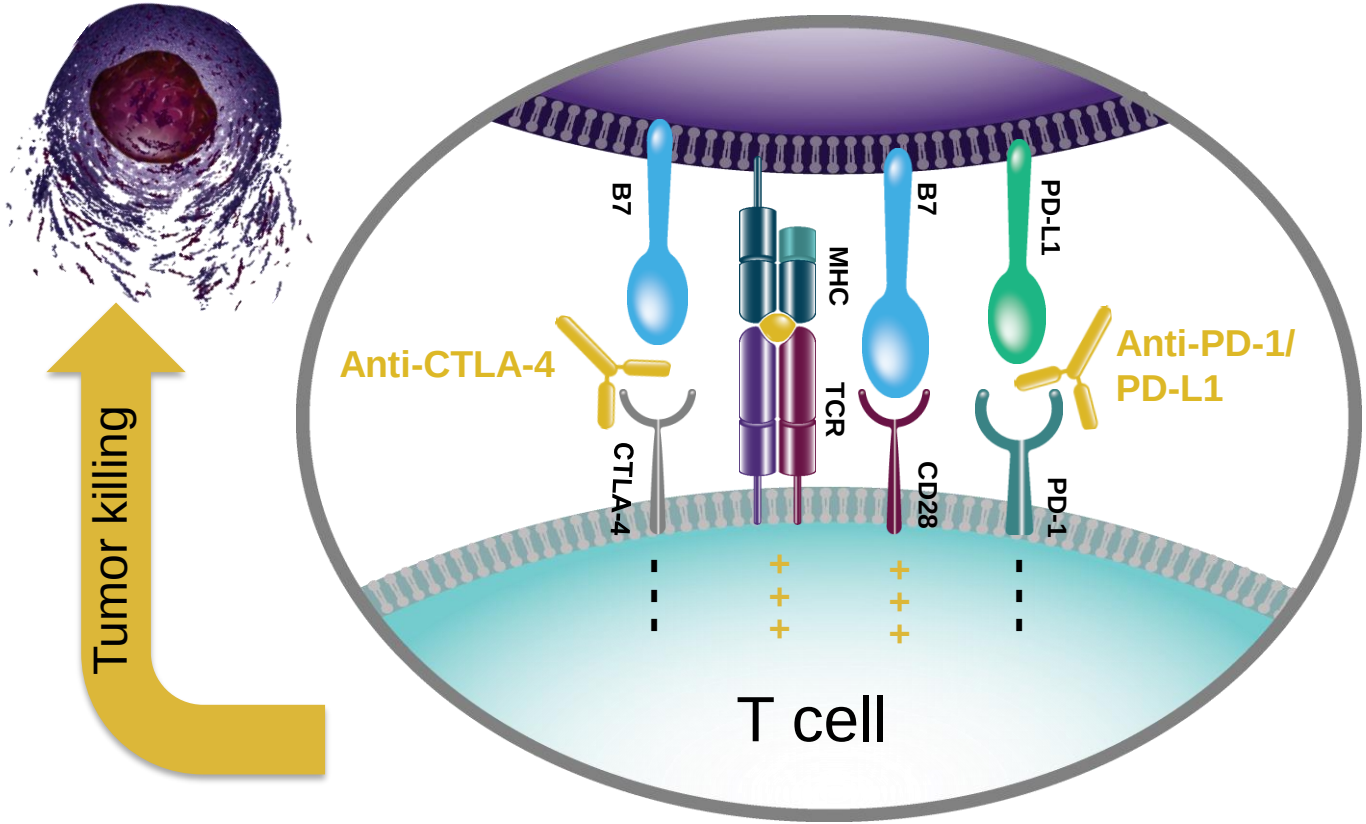
Immune checkpoint blockade is effective in a limited fraction of patients

Ipilimumab Long Term Pooled Survival Analysis:
4846 Patients



Schadendorf, Hodi Wolchok, ESMO, 2013

Inhibition of negative immune regulation



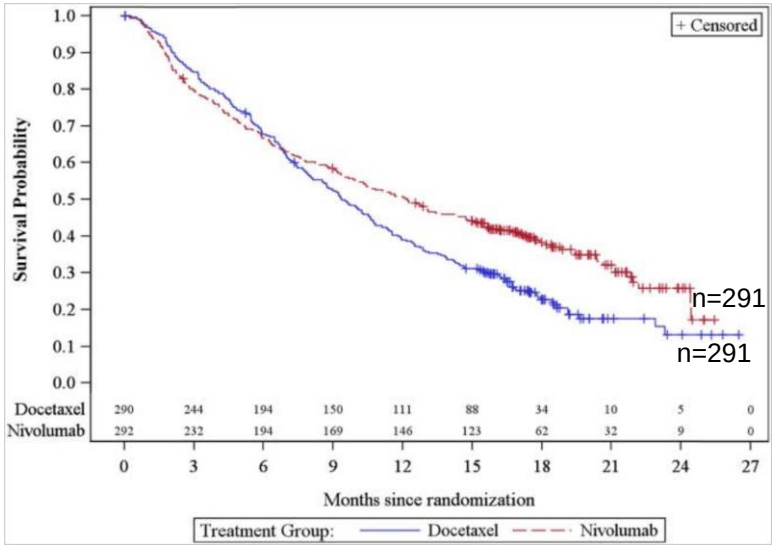
FDA approved anti-CTLA-4
Ipilimumab

FDA approved anti-PD-1
Nivolumab
Pembrolizumab
Cemiplimab

FDA approved anti-PD-L1
Atezolizumab
Avelumab
Durvalumab

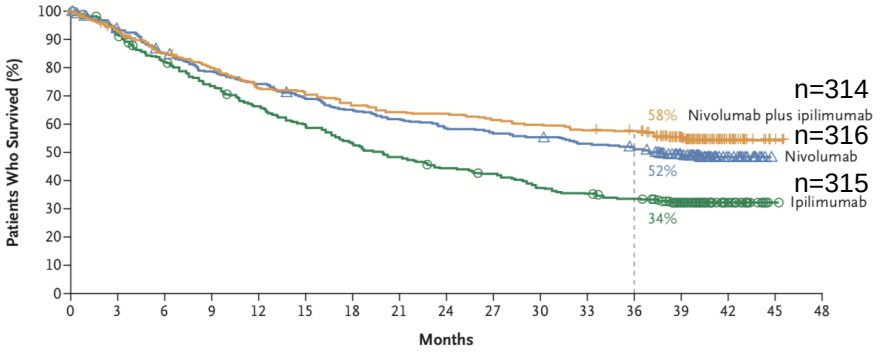
Immune checkpoint blockade is effective in a limited fraction of patients

αPD-1 in advanced NSCLC



CheckMate 057; Kazandjian et al., The Oncologist 2016

αCTLA-4±αPD-1 in advanced melanoma



CheckMate 067
Wolchok et al., NEJM 2017

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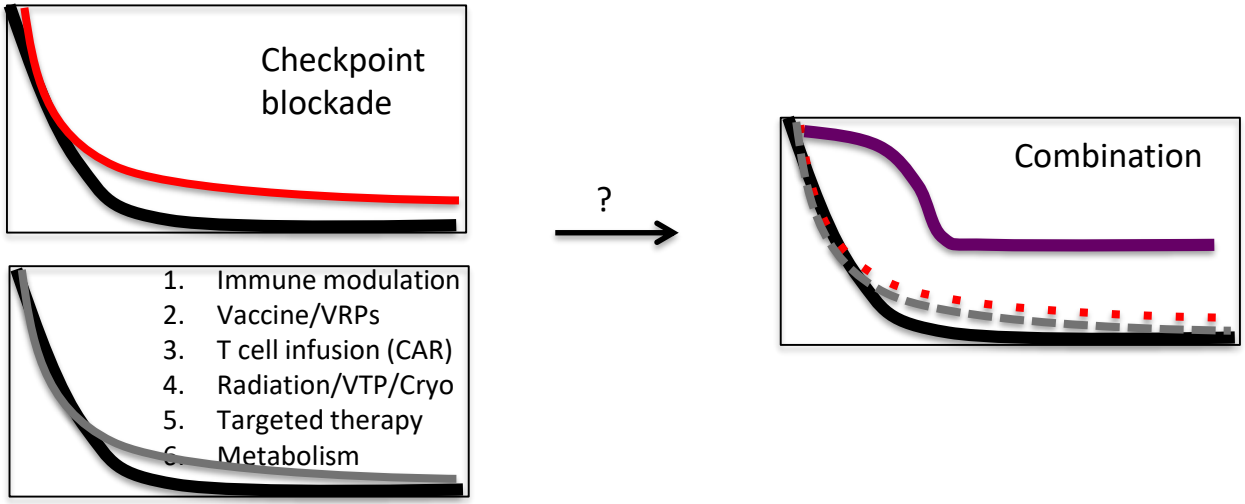
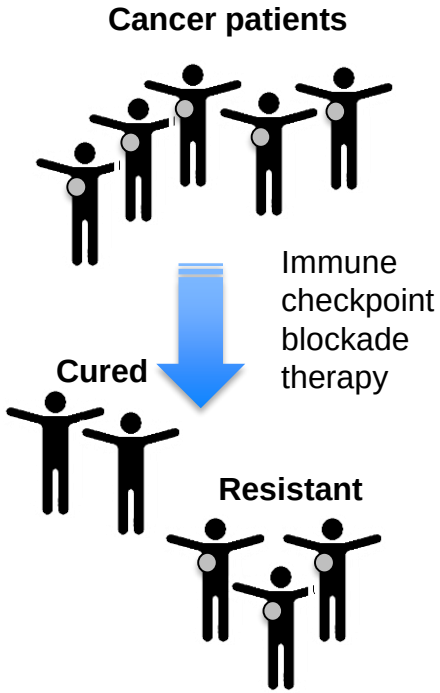
Rationale for Combination with other therapies:

- Use other means to enhance tumor recognition
- Strategy to address low response rates of checkpoint blockade



Lab "focuses" on overcoming major mechanisms of resistance to anti-tumor immunity



Tumor intrinsic resistance

LETTER

doi:10.1038/nature14404

Melanoma-intrinsic β-catenin signalling prevents anti-tumour immunity

Stefani Spranger[1], Riyue Bao[2] & Thomas F. Gajewski[1,3]

Article

Cell

Loss of IFN-γ Pathway Genes in Tumor Cells as a Mechanism of Resistance to Anti-CTLA-4 Therapy

Jianjun Gao,[1,6] Lewis Zhichang Shi,[1,6] Hao Zhao,[5,6] Jianfeng Chen,[1] Liangwen Xiong,[2] Qiuming He,[1] Tenghui Chen,[4] Jason Roszik,[5] Chantale Bernatchez,[5] Scott E. Woodman,[5] Pei-Ling Chen,[5] Patrick Hwu,[5] James P. Allison,[4]

Article

Cell

Genomic and Transcriptomic Features of Response to Anti-PD-1 Therapy in Metastatic Melanoma

Willy Hugo,[1,6,9] Jesse M. Zaretsky,[2,6,9] Lu Sun,[1,6] Chunying Song,[1,6] Blanca Homet Moreno,[2] Siwen Hu-Lieskovan,[2] Beata Berent-Maoz,[2] Jia Pang,[2] Bartosz Chmielowski,[3] Grace Cherry,[2] Elizabeth Seja,[2] Shirley Lomeli,[1,6] Xiangju Kong,[1,6] Mark C. Kelley,[7] Jeffrey A. Sosman,[8] Douglas B. Johnson,[8] Antoni Ribas,[2,3,4,5,6] and Roger S. Lo[1,2,5,6,*]

The NEW ENGLAND JOURNAL of MEDICINE

ESTABLISHED IN 1812 SEPTEMBER 1, 2016 VOL. 375 NO. 9

Mutations Associated with Acquired Resistance to PD-1 Blockade in Melanoma

Jesse M. Zaretsky, B.S., Angel Garcia-Diaz, Ph.D., Daniel S. Shin, M.D., Helena Escuin-Ordinas, Ph.D., Willy Hugo, Ph.D., Siwen Hu-Lieskovan, M.D., Ph.D., Davis Y. Torrejon, M.D., Gabriel Abril-Rodriguez, M.Sc., Salemiz Sandoval, Ph.D., Lucas Barthly, M.Sc., Justin Saco, B.S., Blanca Homet Moreno, M.D., Riccardo Mezzadra, M.Sc., Bartosz Chmielowski, M.D., Ph.D., Kathleen Ruchalski, M.D., I. Peter Shintaku, Ph.D., Phillip J. Sanchez, Ph.D., Cristina Puig-Saus, Ph.D., Grace Cherry, R.N., N.P., Elizabeth Seja, B.A., Xiangju Kong, M.Sc., Jia Pang, B.S., Beata Berent-Maoz, Ph.D., Begoña Comin-Anduix, Ph.D., Thomas G. Graeber, Ph.D., Paul C. Tumeh, M.D., Ton N.M. Schumacher, Ph.D., Roger S. Lo, M.D., Ph.D., and Antoni Ribas, M.D., Ph.D.

Immune-mediated resistance

Cancer Immunol Immunother (2014) 63:247–257
DOI 10.1007/s00262-013-1508-5

ORIGINAL ARTICLE

Frequencies of circulating MDSC correlate with clinical outcome of melanoma patients treated with ipilimumab

Christiane Meyer · Laurène Cagnon · Carla M. Costa-Nunes · Petra Baumgaertner · Nicole Montandon · Loredana Leyvraz · Olivier Michielin · Emanuela Romano · Daniel E. Speiser

Clin Transl Oncol (2016) 18:251–258
DOI 10.1007/s12094-015-1373-0

REVIEW ARTICLE

Tumor-associated macrophages in cancers

W. Hu[1,2,3] · X. Li[1,2,3] · C. Zhang[1] · Y. Yang[1] · L. Jiang[2,3] · C. Wu[1,2,3]

Research article

CTLA4 blockade and GM-CSF combination immunotherapy alters the intratumor balance of effector and regulatory T cells

Sergio A. Quezada, Karl S. Peggs, Michael A. Curran, and James P. Allison

JEM

Article

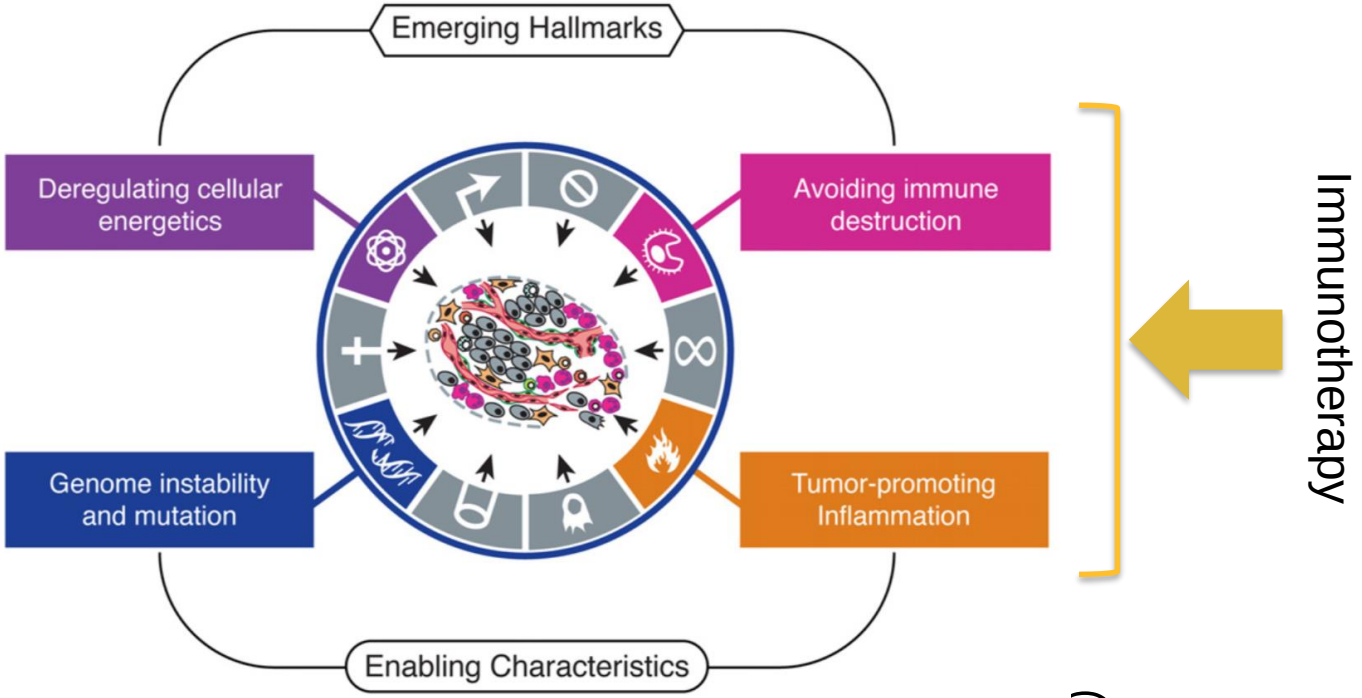
Indoleamine 2,3-dioxygenase is a critical resistance mechanism in antitumor T cell immunotherapy targeting CTLA-4

Rikke B. Holmgaard,[1,2] Dmitriy Zamarin,[1,2,3] David H. Munn,[4] Jedd D. Wolchok,[2,3,5,6] and James P. Allison[1,7]

Hypothesis – Inhibition of tumor glycolysis to overcome resistance to immunotherapy

1. Cellular energy metabolism reprogramming is a critical hallmark of cancer

Inhibition of tumor glycolytic metabolism



Hanahan & Weinberg, Cell 2011

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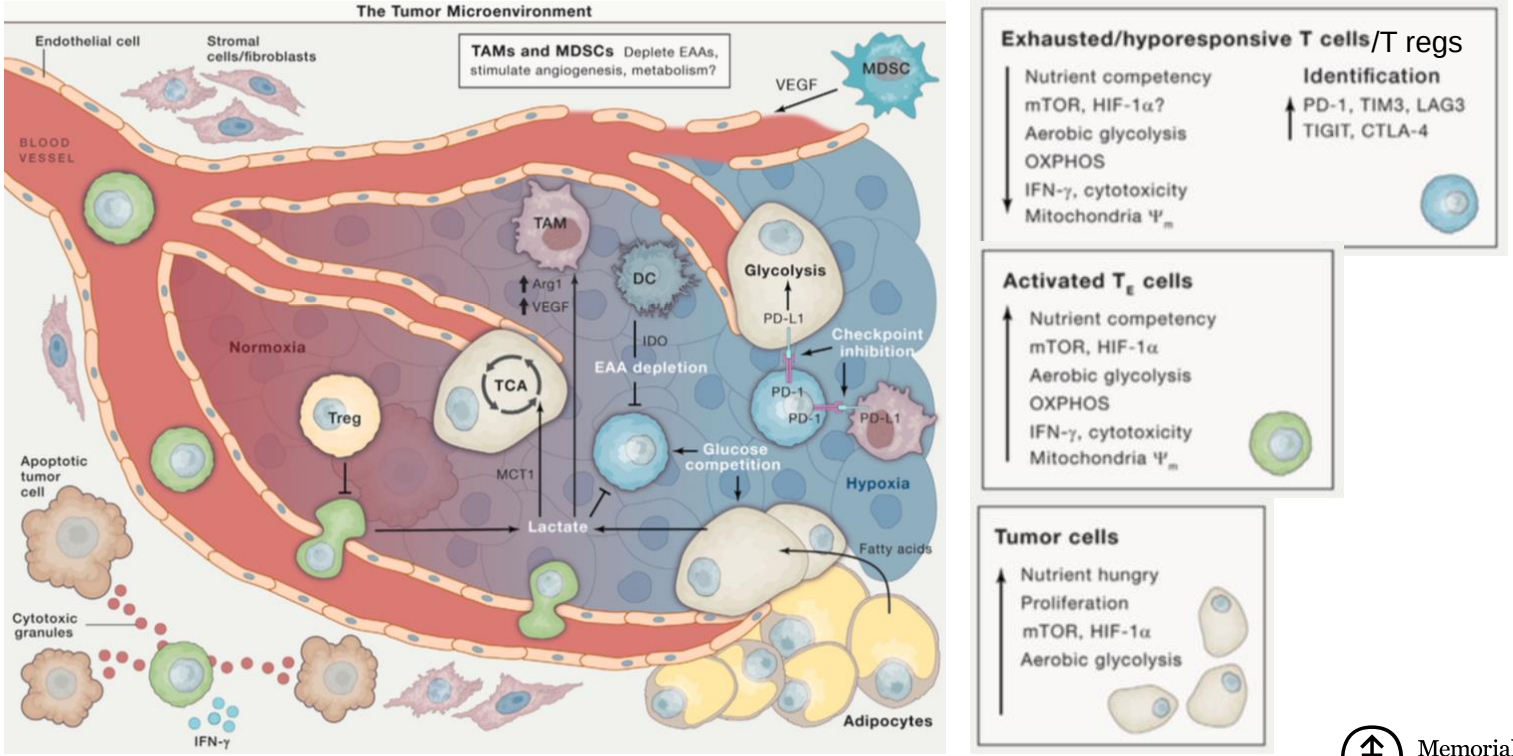
Hypothesis – Inhibition of tumor glycolysis to overcome resistance to immunotherapy

2. Nutrients and oxygen levels within the TME control immune cell function



Buck M.D., Sowell R.T., Kaech S.M., Pearce E.L., Cell 2017

Memorial Sloan Kettering Cancer Center

Hypothesis – Inhibition of tumor glycolysis to overcome resistance to immunotherapy

3. Immune checkpoints and co-stimulatory molecules regulate T cell metabolism

Immunity, Vol. 16, 769–777, June, 2002, Copyright ©2002 by Cell Press

The CD28 Signaling Pathway Regulates Glucose Metabolism

Mitochondrial Priming by CD28

Kenneth A. Frauwirth,[1,2,5] James L. Riley,[1,3,5] Marian H. Harris,[1,2] Richard V. Parry,[1,3] Jeffrey C. Rathmell,[1,2] David R. Plas,[1,2] Rebecca L. Elstrom,[1] Carl H. June,[1,3] and Craig B. Thompson[1,2,4]

Cell

Ramon I. Klein Geltink,[1] David O'Sullivan,[1] Mauro Corrado,[1] Anna Bremser,[2,3] Michael D. Buck,[1] Joerg M. Buescher,[1] Elke Firat,[4] Xuekai Zhu,[5] Gabriele Niedermann,[4,6] George Caputa,[1] Beth Kelly,[1] Ursula Warthorst,[2] Anne Rensing-Ehl,[2] Ryan L. Kyle,[1] Lana Vandersarren,[7,8] Jonathan D. Curtis,[1] Annette E. Patterson,[1] Simon Lawless,[1] Katarzyna Grzes,[1] Jing Qiu,[1] David E. Sanin,[1] Oliver Kretz,[9,10] Tobias B. Huber,[10,11,12] Sophie Janssens[7,8] Bart N. Lambrecht[7,8] Angelika S. Rambold,[2,3] Edward J. Pearce,[1,13] and Erika L. Pearce[1,14,*]

Cell *171*, 385–397, October 5, 2017 © 2017 Elsevier Inc.

ARTICLE

Received 13 Aug 2014 | Accepted 19 Feb 2015 | Published 26 Mar 2015 DOI: 10.1038/ncomms7692 OPEN

PD-1 alters T-cell metabolic reprogramming by inhibiting glycolysis and promoting lipolysis and fatty acid oxidation

Nikolaos Patsoukis[1,2,3], Kankana Bardhan[1,2,3], Pranam Chatterjee[1,2,3], Duygu Sari[1,2,3], Bianling Liu[1,2,3], Lauren N. Bell[4], Edward D. Karoly[4], Gordon J. Freeman[5], Victoria Petkova[1,2,3], Pankaj Seth[2,3,6], Lequn Li[1,2,3] & Vassiliki A. Boussiotis[1,2,3]

4-1BB signaling activates glucose and fatty acid metabolism to enhance CD8+ T cell proliferation

Cellular & Molecular Immunology (2017) 14, 748–757

www.nature.com/cmi

Beom K Choi[1,6], Do Y Lee[1,2,6], Don G Lee[1], Young H Kim[3], Seon-Hee Kim[1], Ho S Oh[1], Chungyong Han[1] and Byoung S Kwon[1,4,5]

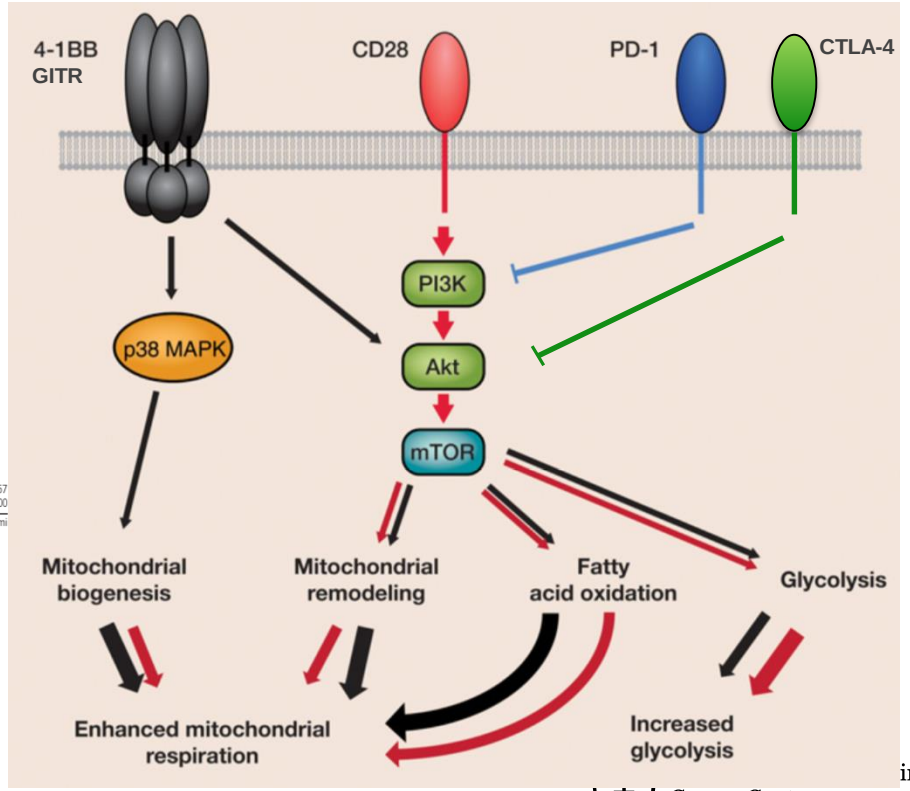
Research Article

Cancer Immunology Research

GITR Agonism Enhances Cellular Metabolism to Support CD8+ T-cell Proliferation and Effector Cytokine Production in a Mouse Tumor Model

Simran S. Sabharwal[1], David B. Rosen[1], Jeff Grein[1], Dana Tedesco[1], Barbara Joyce-Shaikh[1], Roanna Ueda[1], Marie Semana[2], Michele Bauer[2], Kathy Bang[2], Christopher Stevenson[2], Daniel J. Cua[1], and Luis A. Zúñiga[1]

Cancer Immunol Res; 6(10) October 2018



Adapted from Teijeira A. et al, CIR 2019

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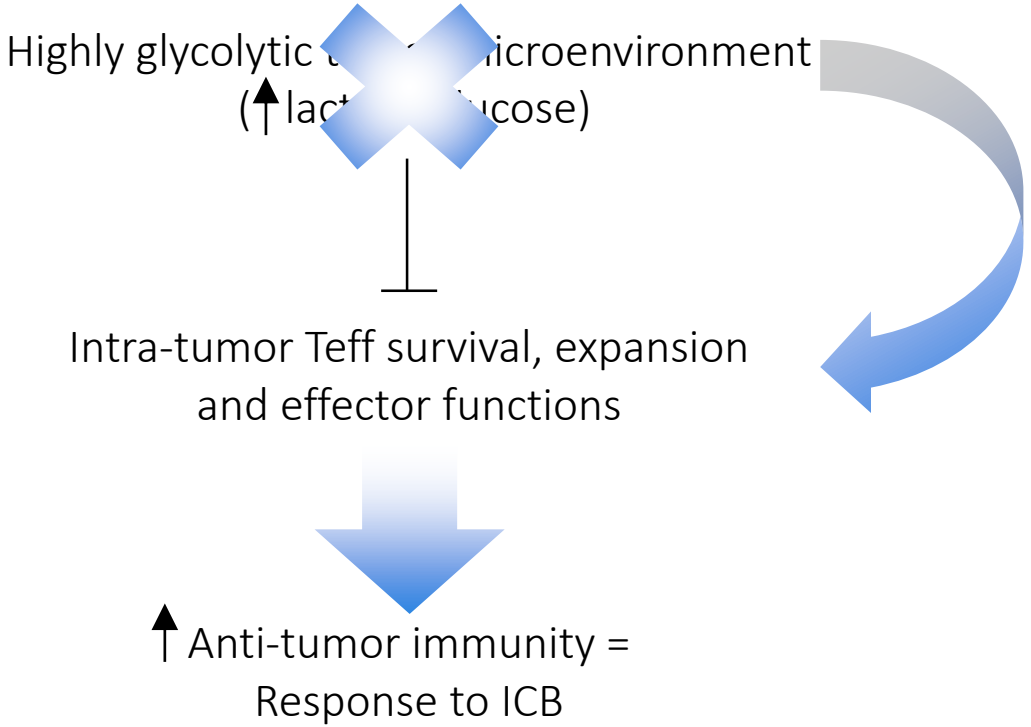
Cancer Center

Working Hypothesis



Aims

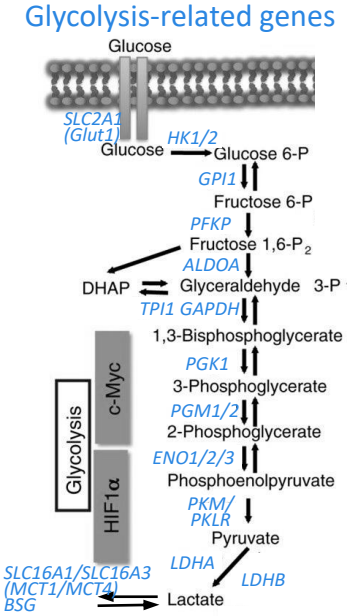
1. Determine how **tumor glycolysis impacts on ICB** activity;

2. Identify the **immune cell types that** are potentiated the most by ICB when tumor glycolysis is hampered;

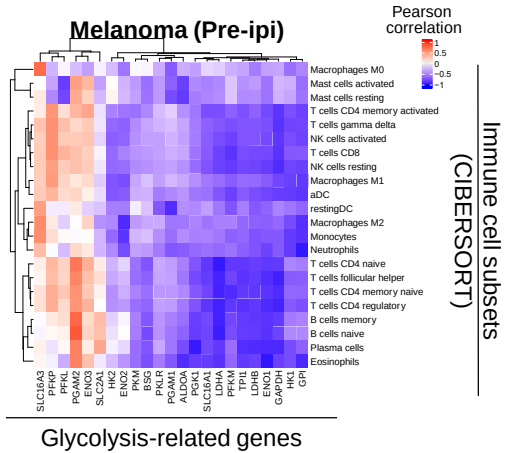
3. Define the **mechanism underlying** these effects.

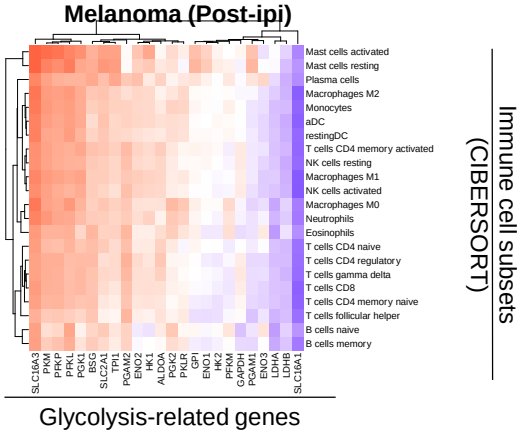
4. Modulate glycolysis **pharmacologically**.

Glycolysis vs. immune infiltration in human tumors before and after immune checkpoint blockade



Heterogeneous tumor glycolysis





Zappasodi et al., *Nature* 2021

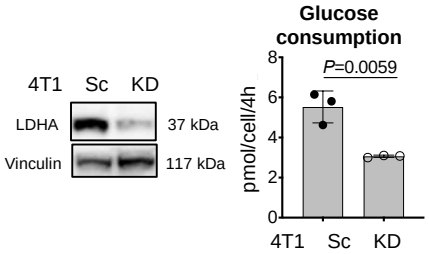
- Immune infiltration and glucose catabolism genes are mutually exclusive;
- CTLA-4 blockade facilitates immune infiltration;
- however, expression of key glycolytic genes and immune infiltrate remain negatively correlated after ipilimumab:

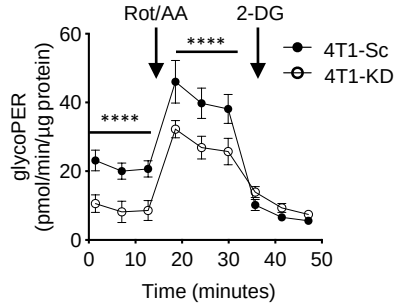
➢ ***Does CTLA-4 blockade work better in glycolysis-low tumors?***

Glycolysis vs. immune infiltration in murine tumors before and after immune checkpoint blockade

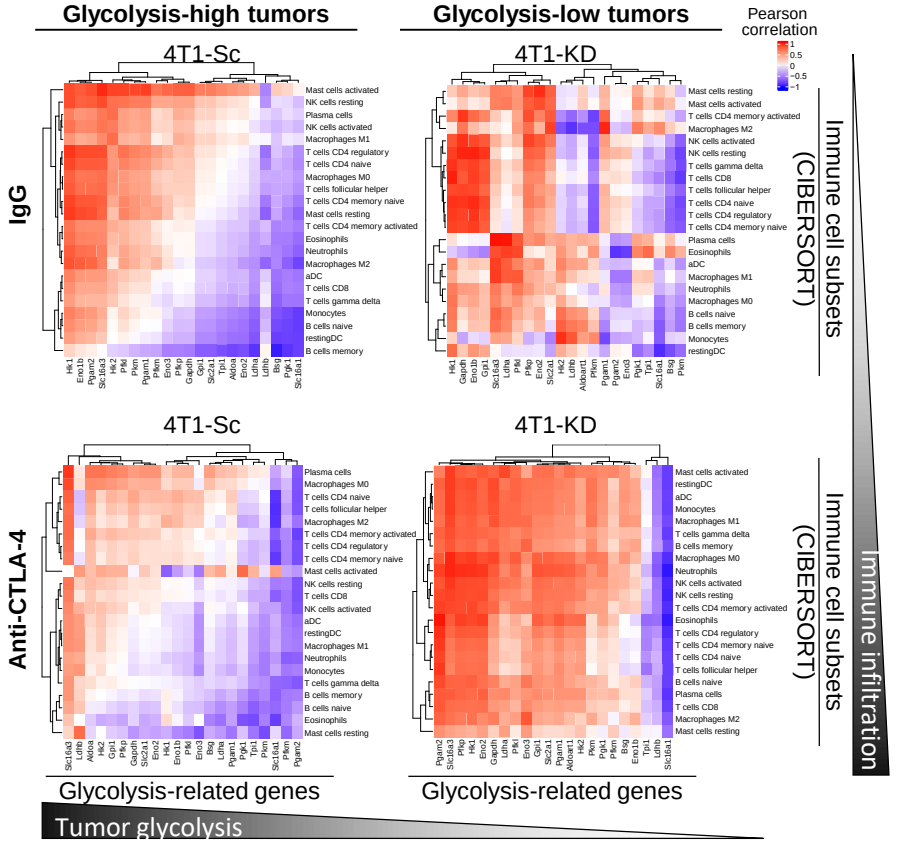
Model System: LDHA knock down in aggressive and immunotherapy resistant mouse tumor models

4T1 mammary carcinoma

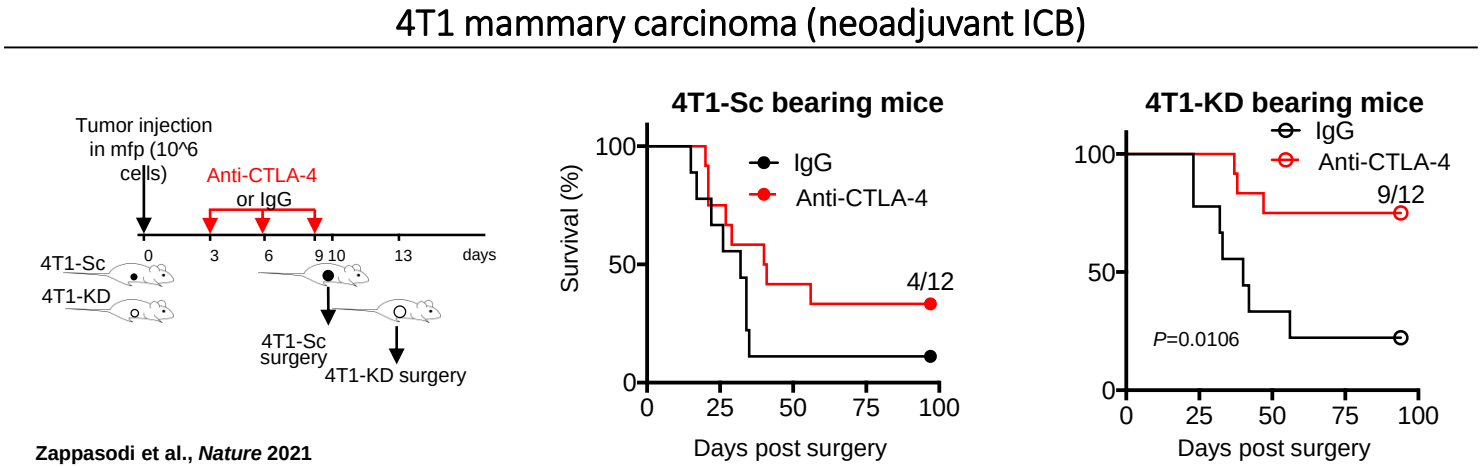




Zappasodi et al., *Nature* 2021



Improved responses of glycolysis-defective tumors to CTLA-4 blockade



- *Increased efficacy and long-lasting anti-tumor immunity of ICB with anti-CTLA-4 in glycolysis-defective LDHA-KD tumors.*
- *Activity of PD-1 blockade is not substantially improved in the same experimental settings.*

Aims

1. Determine how **tumor glycolysis impacts on ICB** activity;

2. Identify the **immune cell types that** are potentiated the most by ICB when tumor glycolysis is hampered;

3. Define the **mechanism underlying** these effects.

4. Modulate glycolysis pharmacologically.

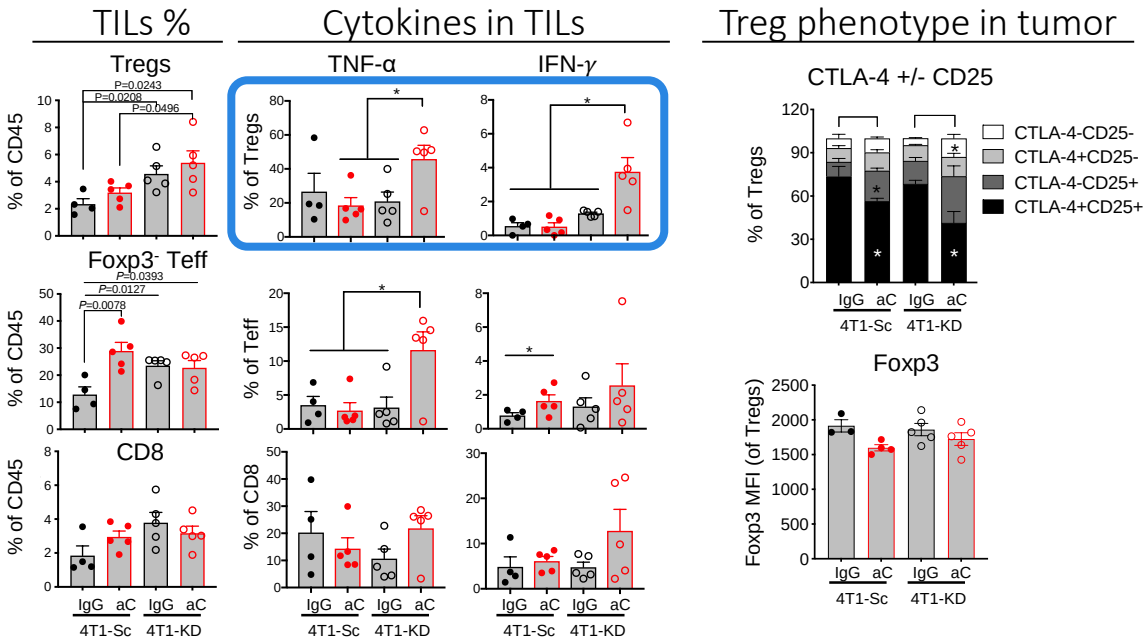
CTLA-4 blockade drives in Treg instability in glycolysis-defective tumors

- Changes in frequency of TIL subsets: NO

✓ Changes in Treg function: YES*

✓ Changes in Treg phenotype: YES*

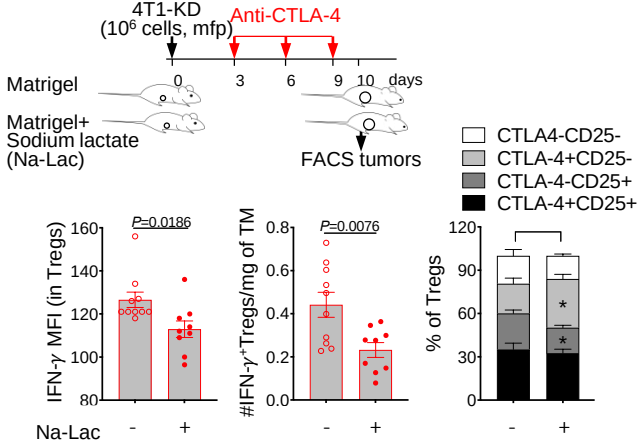
**Similar results with B16-KD vs. B16-Sc*



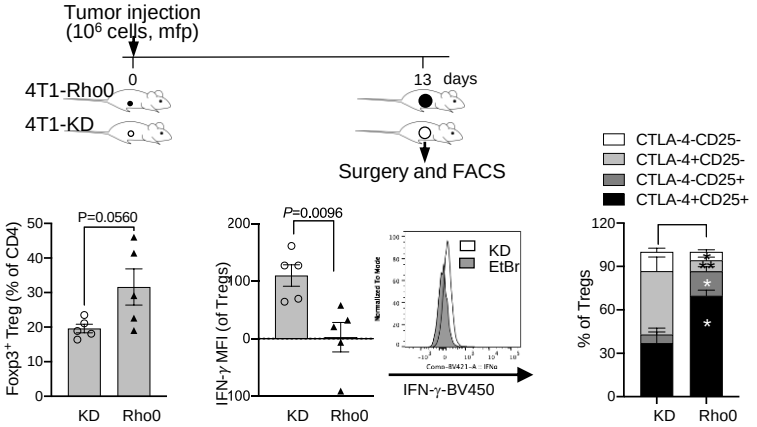
Zappasodi et al., *Nature* 2021

Does the local lactate:glucose ratio play a role?

Supplying lactate in the KD TME



Maximizing tumor glycolysis (4T1-Rho0)



Zappasodi et al., *Nature* 2021

- *Increasing lactate:glucose ratio or boosting tumor glycolysis potentiates Treg stability;*
- *Loss of Treg stability induced by anti-CTLA-4 in glycolysis-defective tumors is* ***dependent on local lactate:glucose ratio.***

Aims

1. Determine how **tumor glycolysis impacts on ICB** activity;

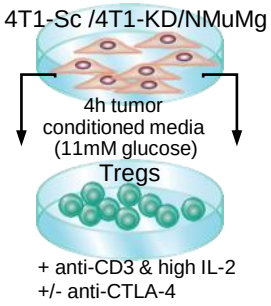
2. Identify the **immune cell types** that are potentiated the most by ICB when tumor glycolysis is hampered;

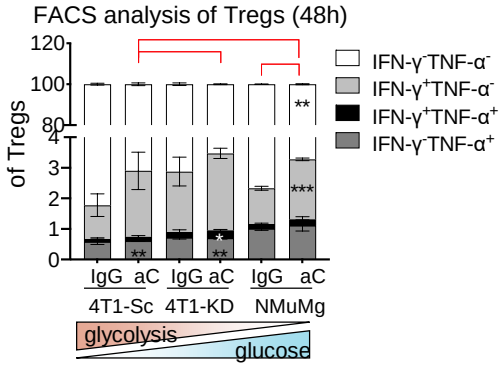
3. Define the **mechanism underlying** these effects.

4. Modulate glycolysis **pharmacologically.**

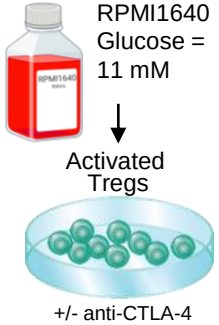
Anti-CTLA-4 promotes Treg glucose utilization and IFN-γ production associated with reduced Treg suppression

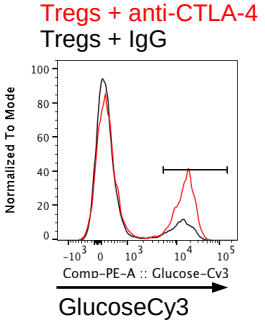
Tregs in tumor conditioned media ± anti-CTLA-4

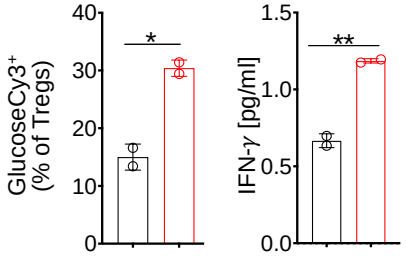




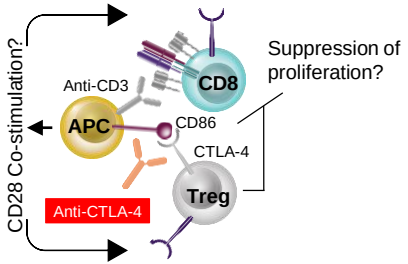
Tregs cultured alone ± anti-CTLA-4

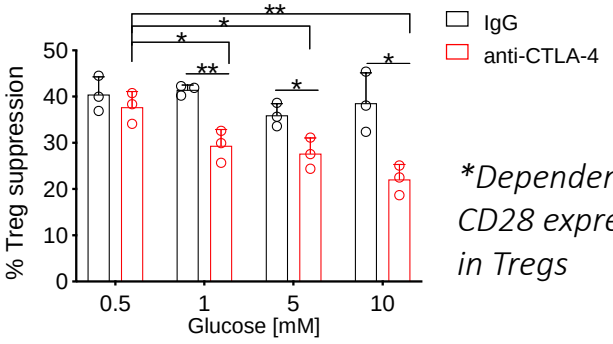






Treg suppression assay in increasing [glucose] ± anti-CTLA-4



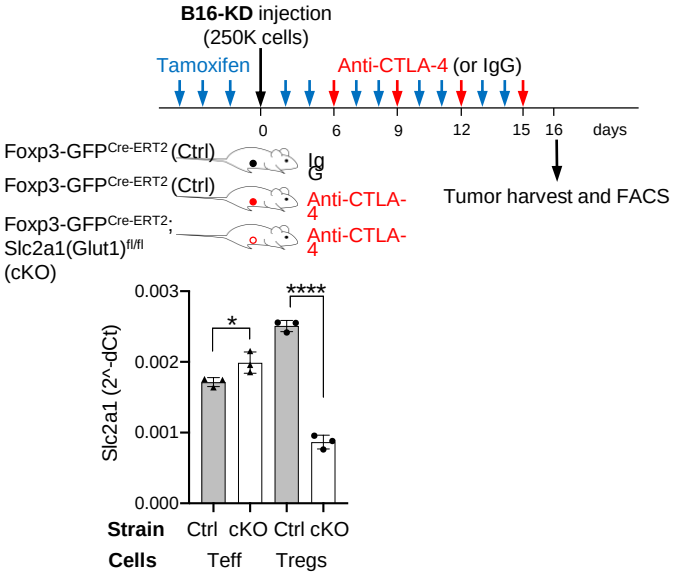


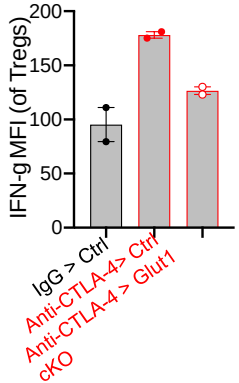
Dependent on CD28 expression in Tregs

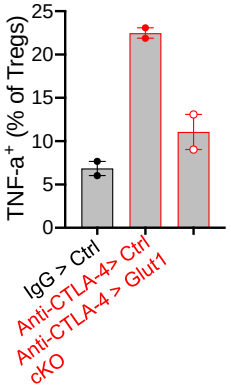
Zappasodi et al., *Nature* 2021

Loss of Treg stability upon anti-CTLA-4 is dependent on Treg capacity to metabolize glucose *in vivo*

LDHA-KD tumors (↑**glucose**:lactate) → Glut1 cKO mice (no glucose uptake in Tregs) --Anti-CTLA-4 (or IgG)--> IFN-γ, TNF-α production by Tregs?

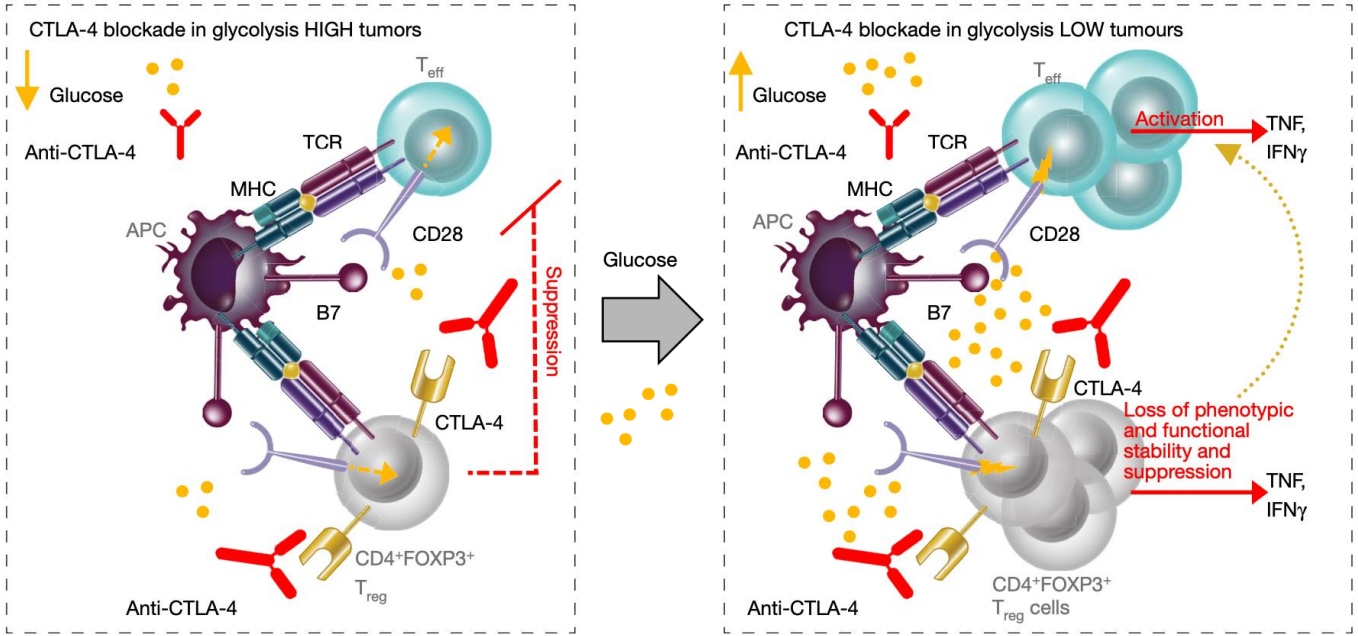






Zappasodi et al., *Nature* 2021

Model & Perspectives



CTLA-4 blockade may be best exploited to treat glycolysis-low tumors and/or in combination with inhibitors of tumor glycolysis.

Aims

1. Determine how **tumor glycolysis impacts on ICB** activity;

2. Identify the **immune cell types that** are potentiated the most by ICB when tumor glycolysis is hampered;

3. Define the **mechanism underlying** these effects.

4.Modulate glycolysis **pharmacologically.**

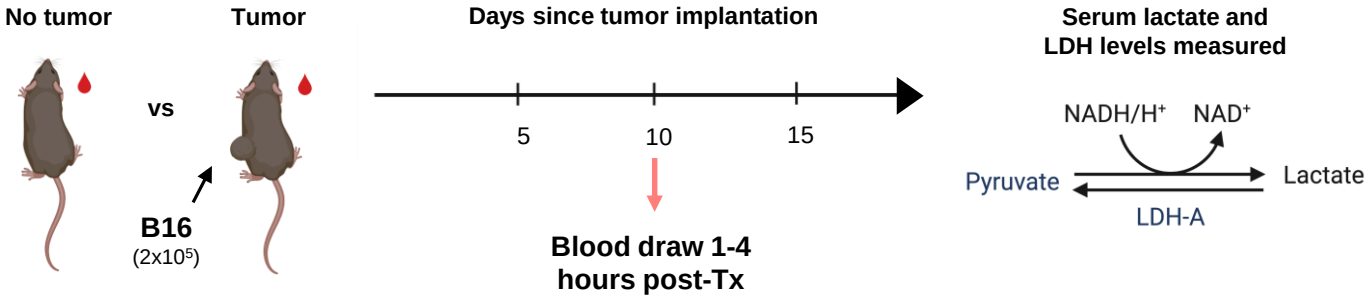
Pharmacological Modulation of Metabolism to Improve ICB

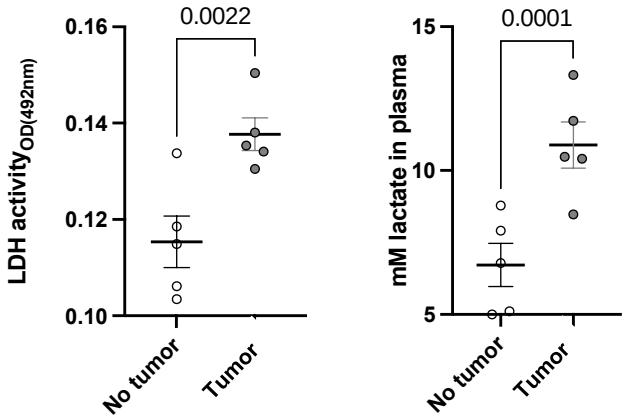
- Effects of LDH pharmacologic inhibition
- Explore the effects Lactate transport

Pharmacological Modulation of Metabolism to Improve ICB

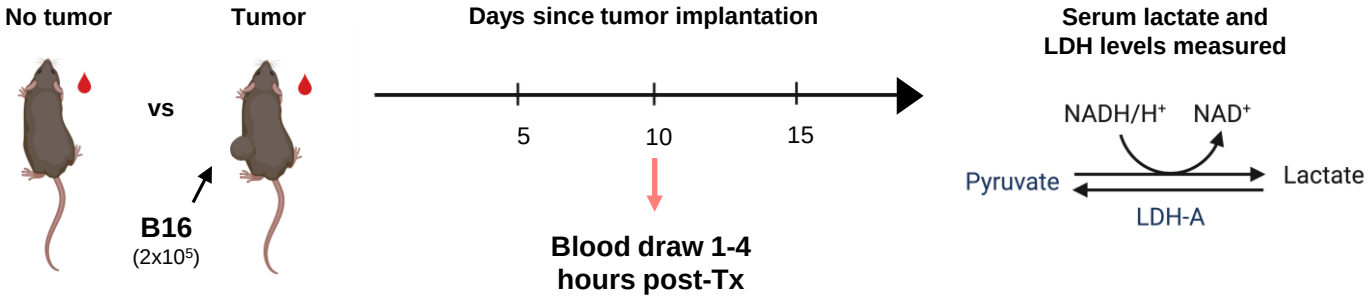
- Effects of LDH pharmacologic inhibition
- Explore the effects Lactate transport

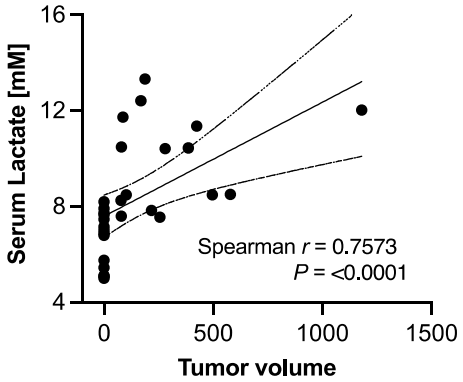
Mice bearing B16 melanoma tumors have higher serum LDH and lactate levels than non-tumor bearing mice

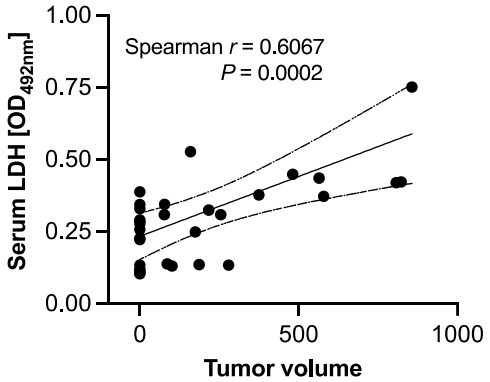




Serum lactate and LDH levels correlate with B16 melanoma tumor burden





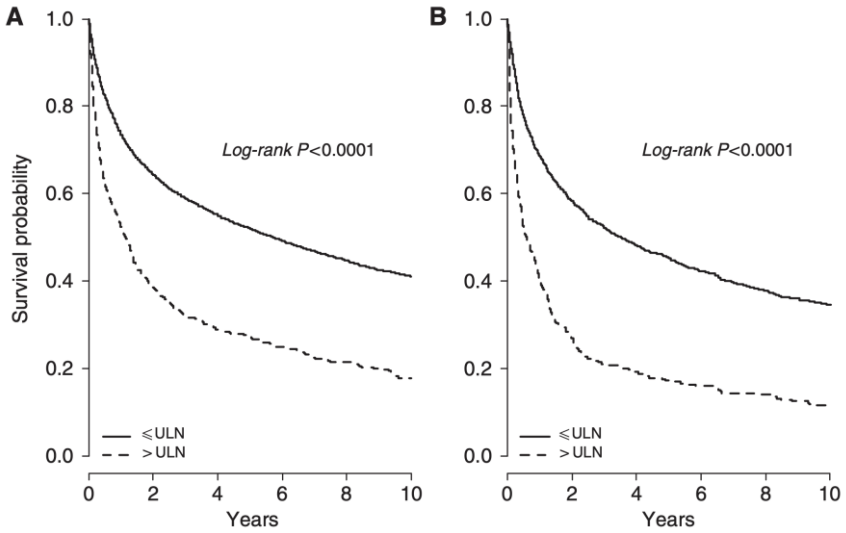


High serum LDH levels are a negative prognostic factor for many cancers

Patients with serum LDH levels above baseline prior to diagnosis have a lower survival probability

Serum lactate dehydrogenase and survival following cancer diagnosis

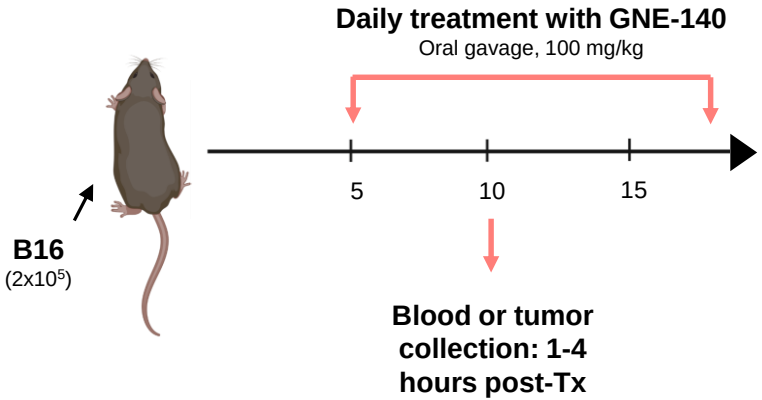
Wahyu Wulaningsih*[,1], Lars Holmberg[1,2,3], Hans Garmo[1,3], Håkan Malmstrom[4], Mats Lambe[3,5], Niklas Hammar[4,6], Göran Walldius[7], Ingmar Jungner[8], Tony Ng[9] and Mieke Van Hemelrijck[1,4]

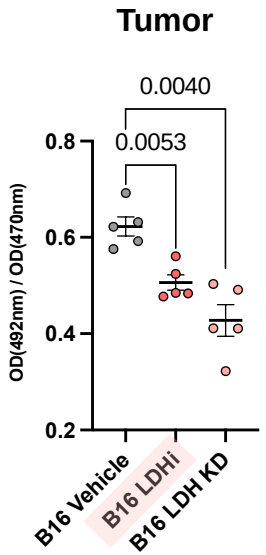


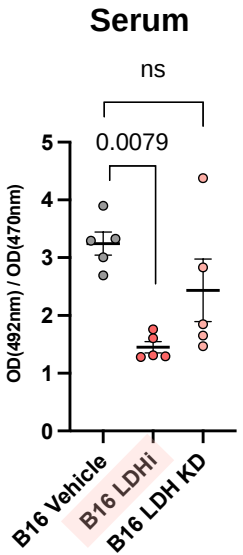
Kaplan-Meier curves for 10-year overall survival following cancer diagnosis by serum LDH (above or below baseline levels) measured within (**A**) 3 years before diagnosis and (**B**) 3 months before diagnosis.

In vivo: When administered daily, GNE-140 (LDHi) reduces LDH activity within the tumor and in the periphery

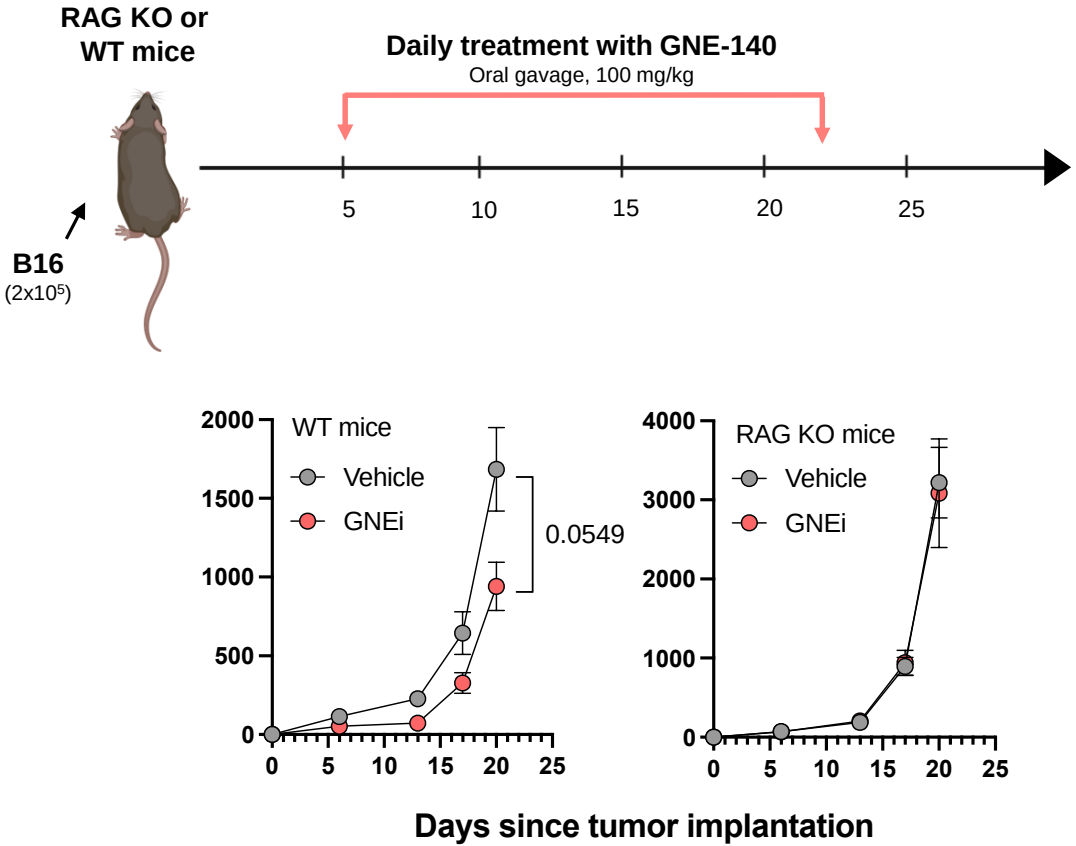
Mice bearing LDHA KD tumors have low tumor LDH but not significantly lower serum LDH





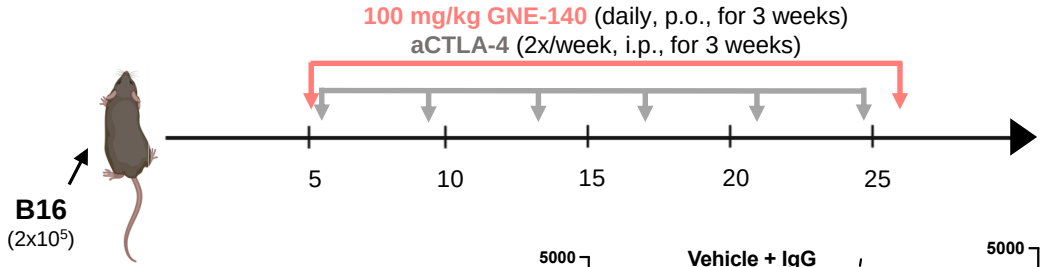


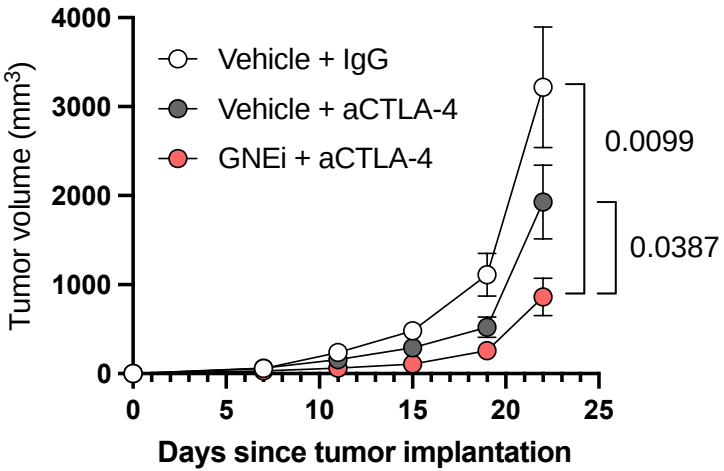
Adaptive immunity is required for the anti-tumor effect of LDHi

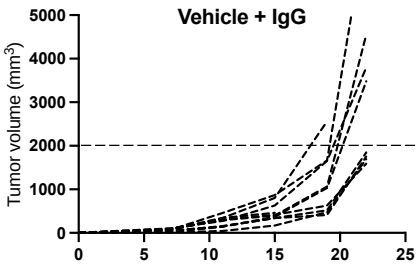


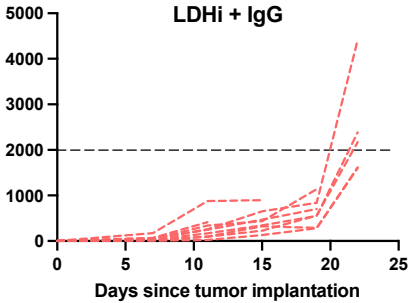
n = 10 mice/group

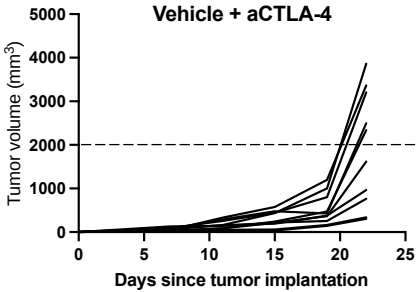
Treatment with LDHi sensitizes B16 melanoma to CTLA-4 blockade

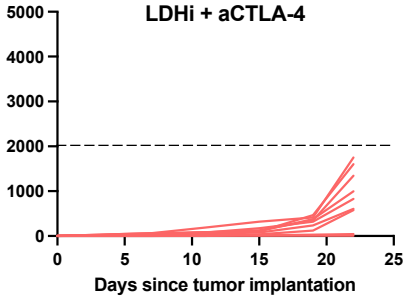








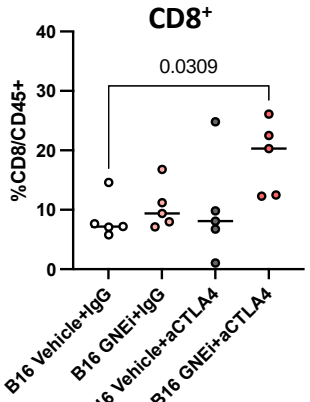


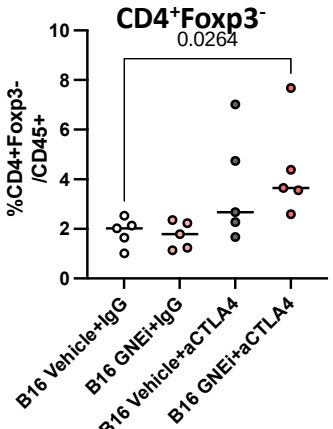


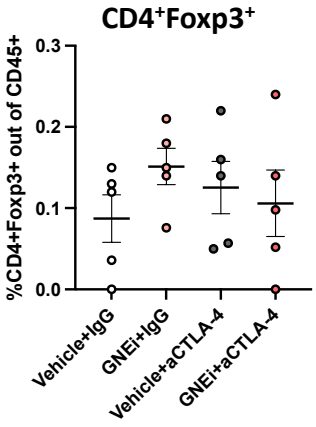
n = 10 mice/group

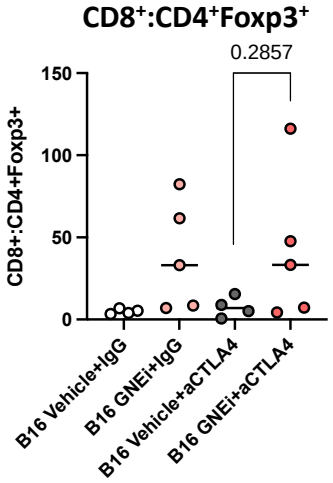
CTLA-4 blockade combined with LDH inhibition leads to increased $CD8^+$ and $CD4^+$ effector infiltration without increasing Treg infiltration

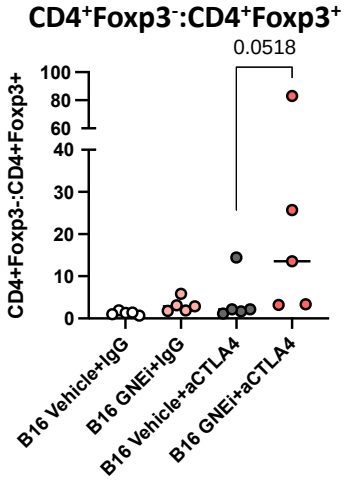
Immune infiltrate was examined after the 3rd anti-CTLA-4 administration, 2 weeks post-tumor implantation







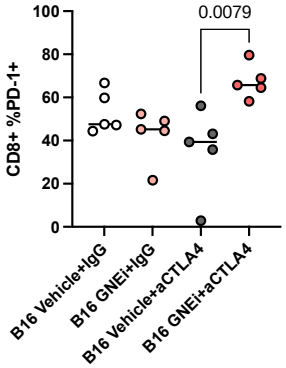


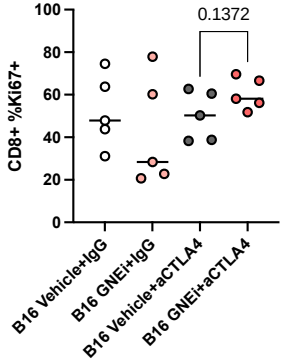


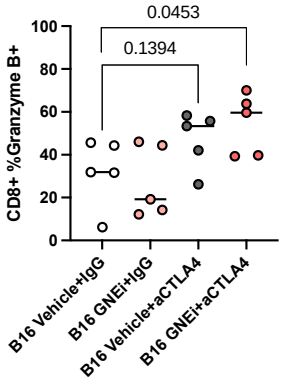
LDHi combined with CTLA-4 blockade favors CD8⁺ and CD4⁺ effector activation

LDHi may be preferentially targeting tumor cells that overexpress LDH, alleviating competition for glucose in the tumor microenvironment

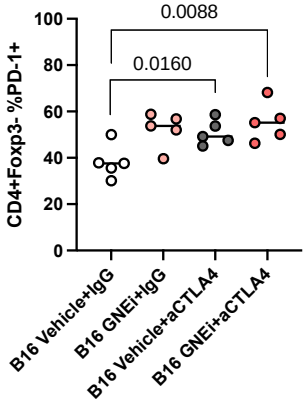
CD8⁺ phenotypes

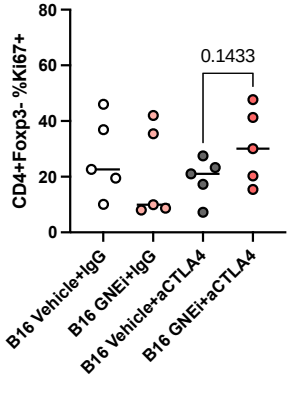


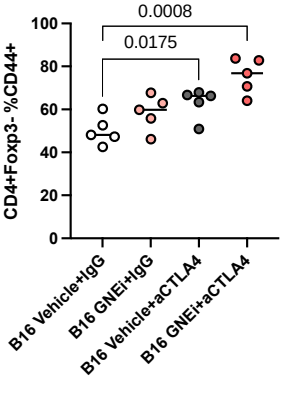




CD4⁺Foxp3⁻ phenotypes



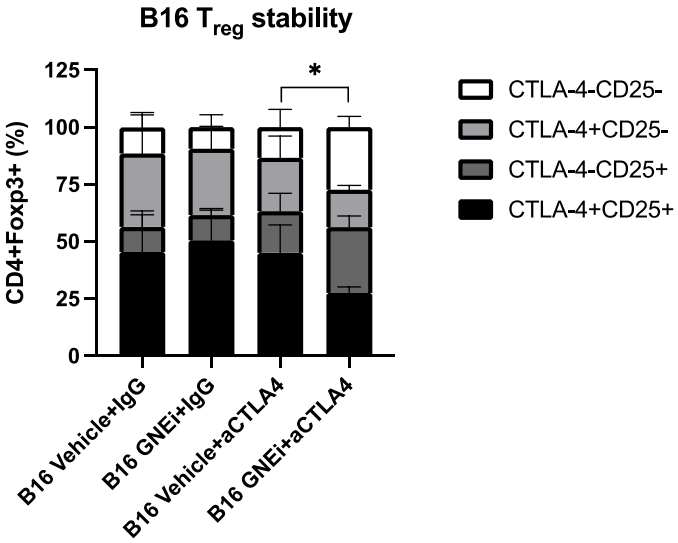


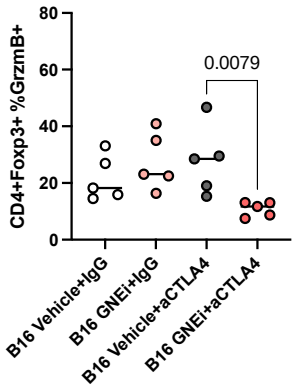


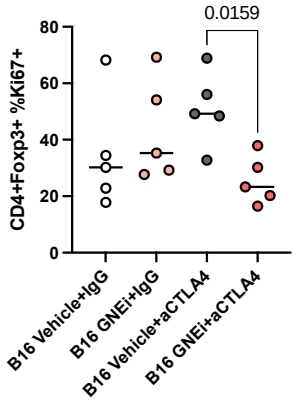
Similarly to when LDH is downregulated within the tumor, LDH inhibition leads to loss of Treg stability in the setting of CTLA-4 blockade

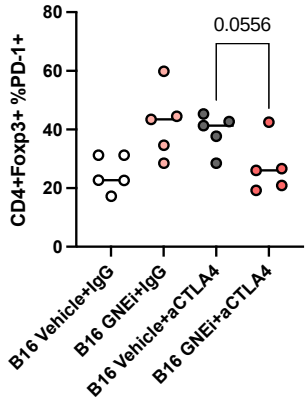
We had previously shown that LDHA-deficient Tregs displayed poor expansion and proliferation potential post-treatment with CTLA-4 blockade

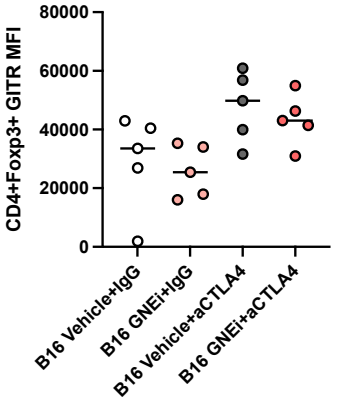
CD4⁺Foxp3⁺ phenotypes







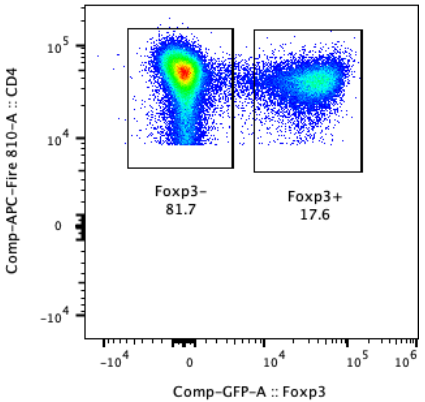




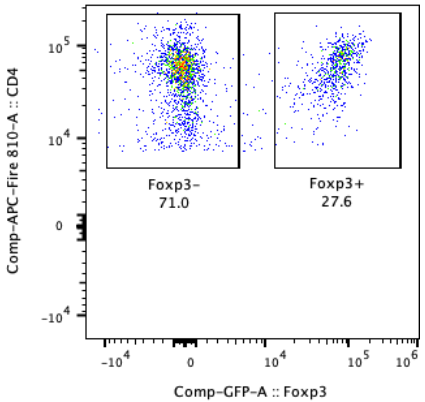
Treatment with LDHi increases glucose uptake capacity of tumor-infiltrating T cells, including Foxp3⁺ regulatory T cells, but not splenocytes

Foxp3 GFP mice:

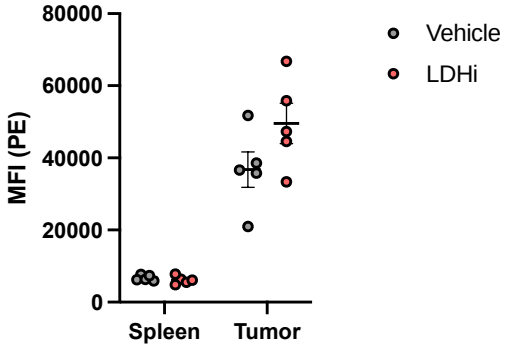
Spleen



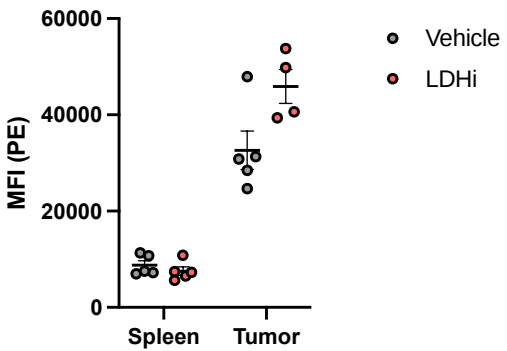
Tumor



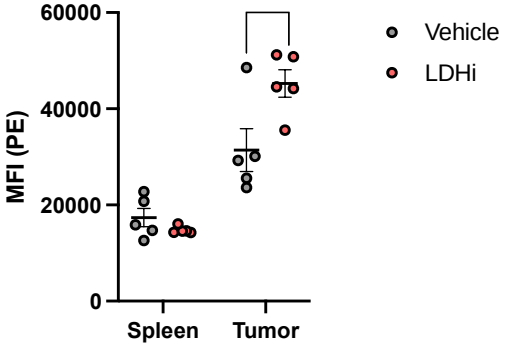
CD8+ Glucose Cy3 MFI



CD4+Foxp3- Glucose Cy3 MFI



CD4+Foxp3+ Glucose Cy3 MFI



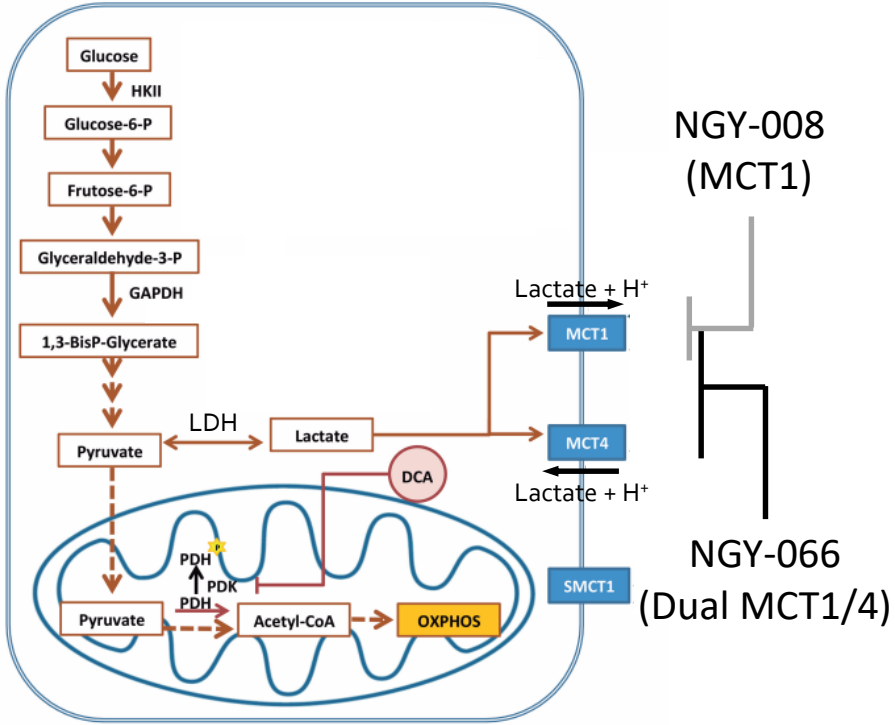
Conclusions

- Serum lactate and LDH levels correlate with B16 melanoma tumor burden, and patients with serum LDH levels above baseline prior to diagnosis have a lower survival probability
- LDH inhibitor GNE-140 reduces tumor cell glycolysis in vitro, lowers tumor and serum LDH in mice, and delays B16 melanoma growth in immunocompetent but not immunodeficient mice
- Treatment with LDHi alongside CTLA-4 blockade delays tumor growth more significantly than immunotherapy alone
- CTLA-4 blockade combined with LDH inhibition leads to increased $CD8^+$ and $CD4^+$ effector infiltration and activation, while resulting in functional destabilization of regulatory T cells
- Upon treatment with LDHi *in vivo,* tumor-infiltrating $CD8^+$, $CD4^+Foxp3^-$, and $CD4^+Foxp3^+$ display a higher capacity for glucose uptake, while splenocyte glucose uptake remains unchanged, indicating that LDHi is relatively tumor-specific, due to the tumor's over-reliance on glycolysis and overexpression of LDH

Pharmacological Modulation of Metabolism to Improve ICB

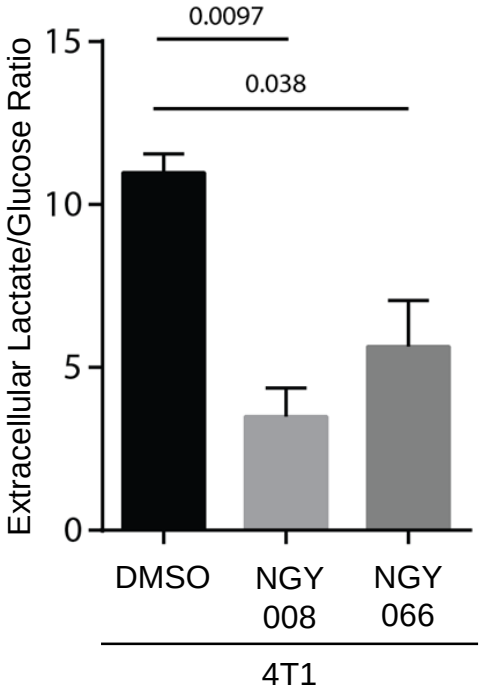
- Effects of LDH pharmacologic inhibition
- Explore the effects Lactate transport

Inhibition of MCTs reduces tumor lactate production



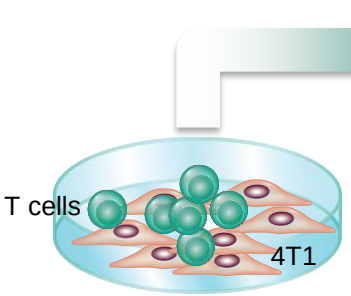
NGY-008 (MCT1)

NGY-066 (Dual MCT1/4)



Inhibition of MCTs favors T-cell activation in tumor-T-cell co-cultures

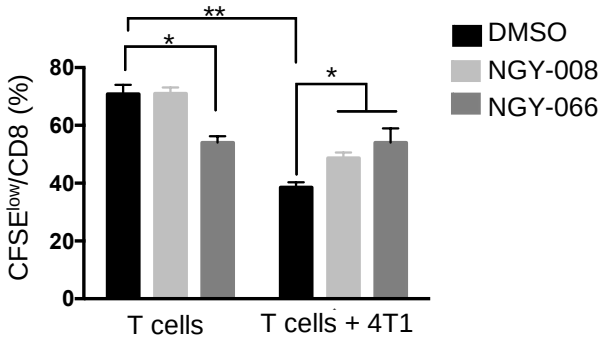
CFSE-labeled T cells



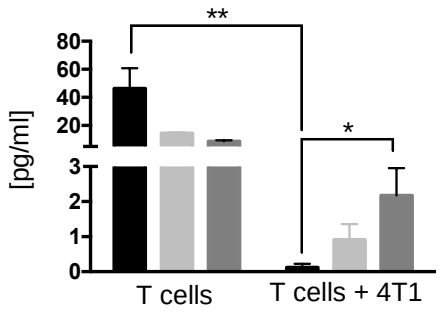
+PHA or anti-CD3/CD28
+vehicle/008/066

Culture supernatants

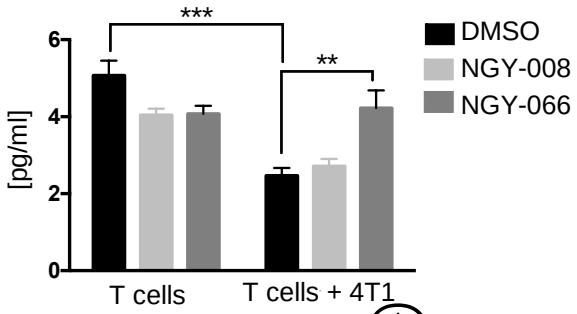
Proliferation



IFN-γ



TNF-α



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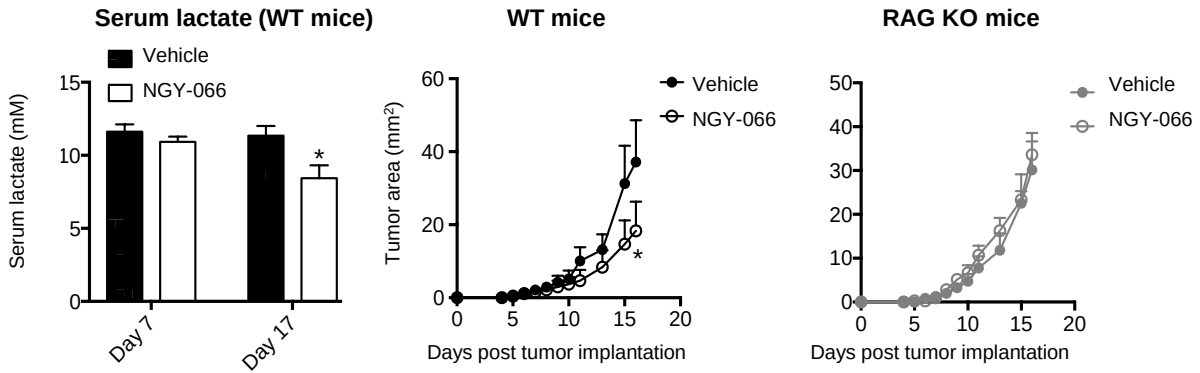
Immune mediated effects of pharmacologic modulation of tumor lactate metabolism *in vivo*



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Conclusions

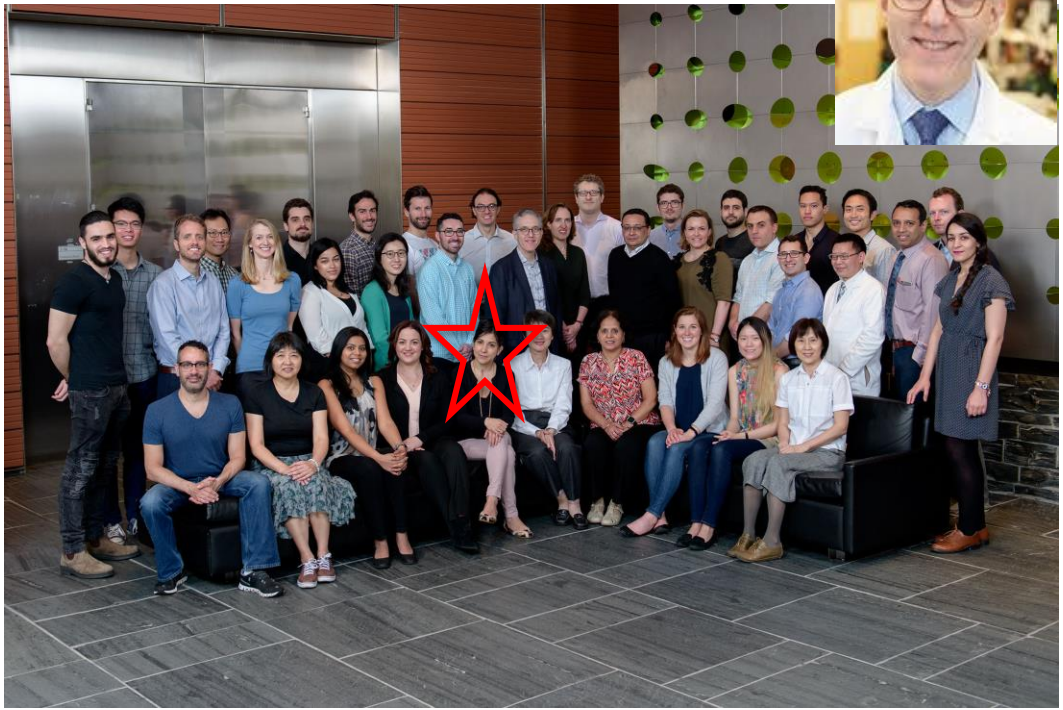
1. Co-culture of T cells with 4T1 cells significantly limits their immune effector functions

2. Blocking lactate transporters *in vitro* improves T-cell functions

3. Inhibition of lactate transport *in vivo* delays tumor progression in a manner that is dependent on adaptive immunity

→ Explore MCT expression patterns in different model systems

Acknowledgments



Ludwig Collaborative and Swim Across America Lab @ MSK
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Mario Colombo (Milan)

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University of Pittsburgh

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Thank you!

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