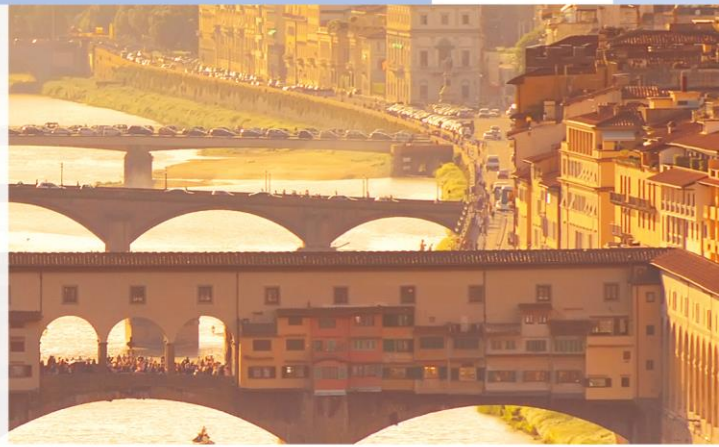
8° WORKSHOP IN EMATOLOGIA TRASLAZIONALE

DELLA SOCIETÀ ITALIANA DI EMATOLOGIA SPERIMENTALE

Firenze - Auditorium CTO - A.O.U. Careggi, 22-23 giugno 2023



Alterazioni del microambiente e del metabolismo: alleati o punti deboli della leucemia mieloide acuta?

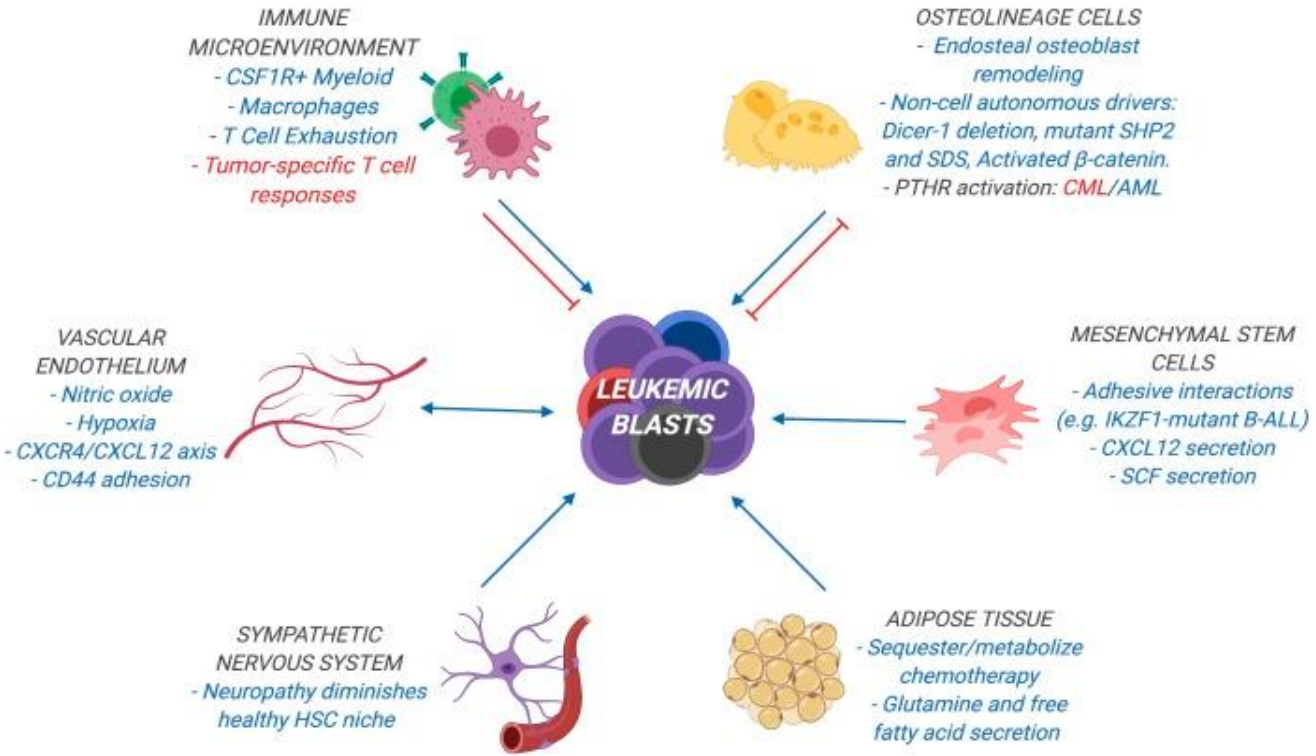
Antonio Curti

IRCCS Azienda Ospedaliero-Universitaria di Bologna

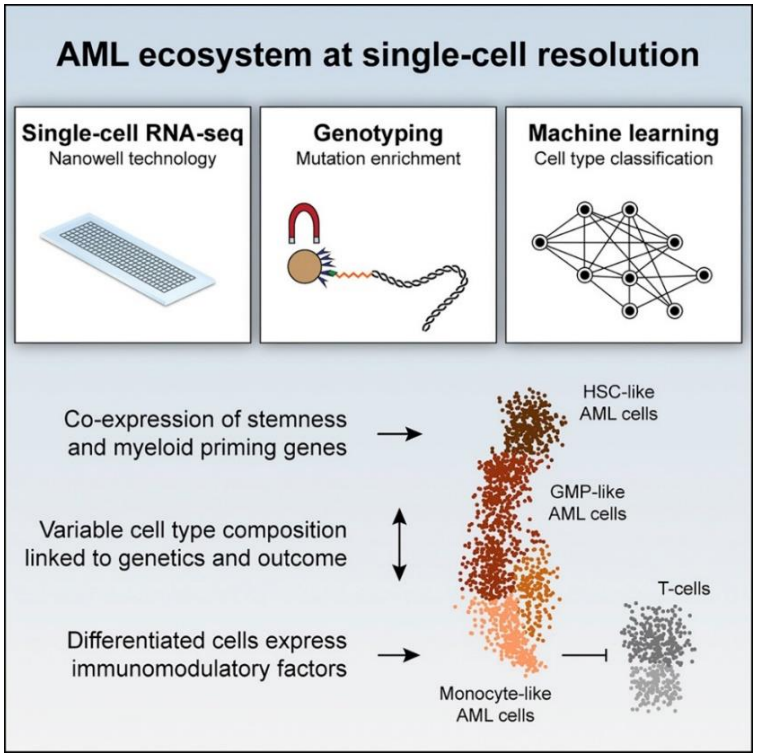
Istituto di Ematologia «Seràgnoli», Bologna

Disclosures of Name Surname

Company name	Research support	Employee	Consultant	Stockholder	Speakers bureau	Advisory board	Other
Abbvie	X					X	
Novartis						X	
Pfizer						X	
Jazz Pharma						X	
Servier						X	



Witkowski MT, et al. J Exp Med. 2020



Single-Cell RNA-seq reveals AML hierarchies relevant to disease progression and immunity

Recent studies using single cell sequencing have revealed the clonal diversity and phenotypic heterogeneity in AML with greater precision.

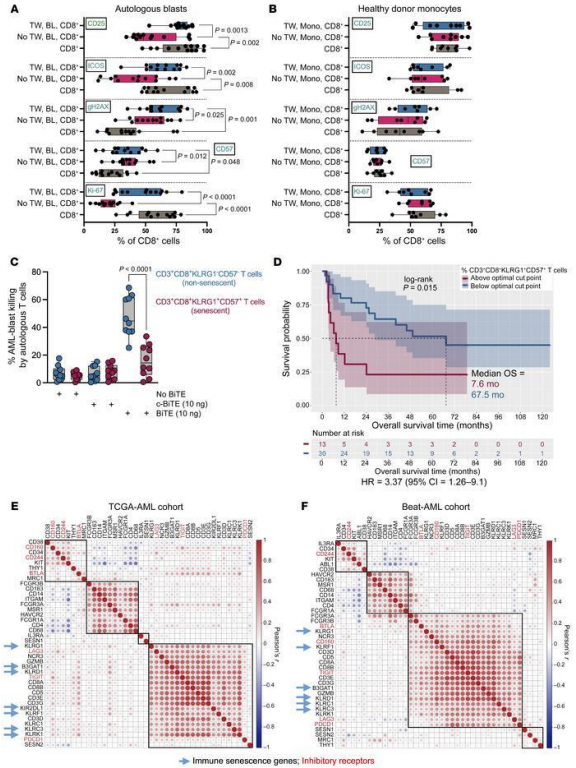
Cell ontogeny and function of leukemic cells may impact T cell responses, as single-cell sequencing revealed that monocytic AML cells are associated with more suppressive T cell landscapes.

Van Galen P, Cell, Volume 176, 2019, Pages 1265-1281

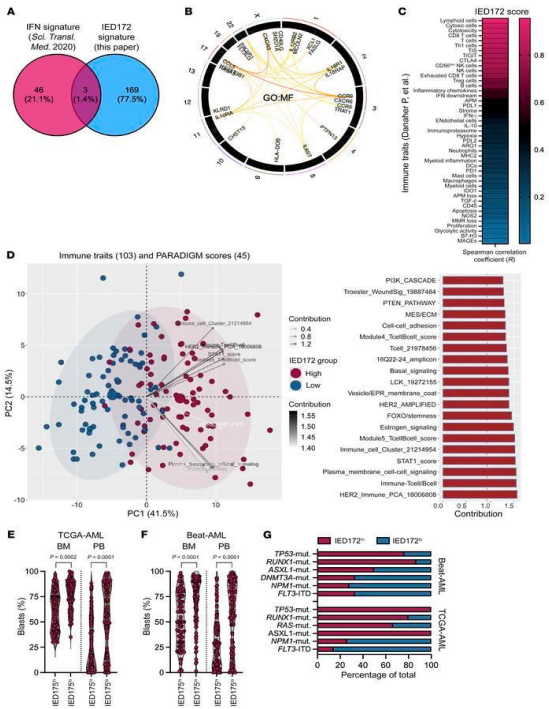
The immune landscape in AML: major issues

1. Increased Treg cell number
2. Increased T cell exhaustion, such as through upregulation of immune checkpoint ligands and receptors, and senescence
3. Diminished function of T helper and alteration in cytokine production
4. Deregulated anti-leukemic NK-mediated cytotoxicity
5. Increased myeloid derived suppressor cell and M2-like macrophage populations

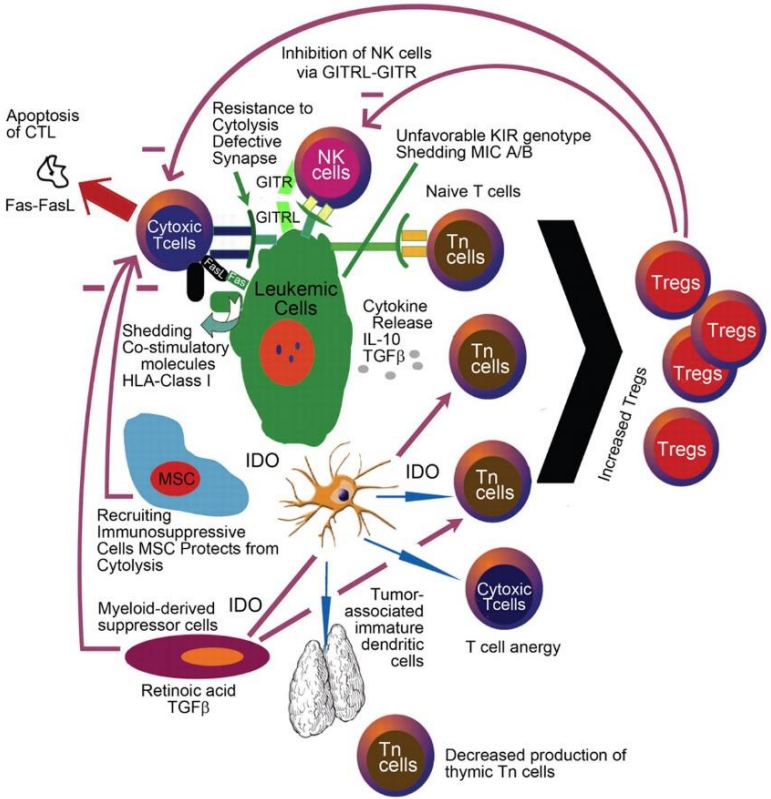
Markers of T cell senescence correlate with impaired T cell killing and poor clinical outcomes



Signatures of immune effector dysfunction correlate with immune infiltration and with adverse-risk molecular features



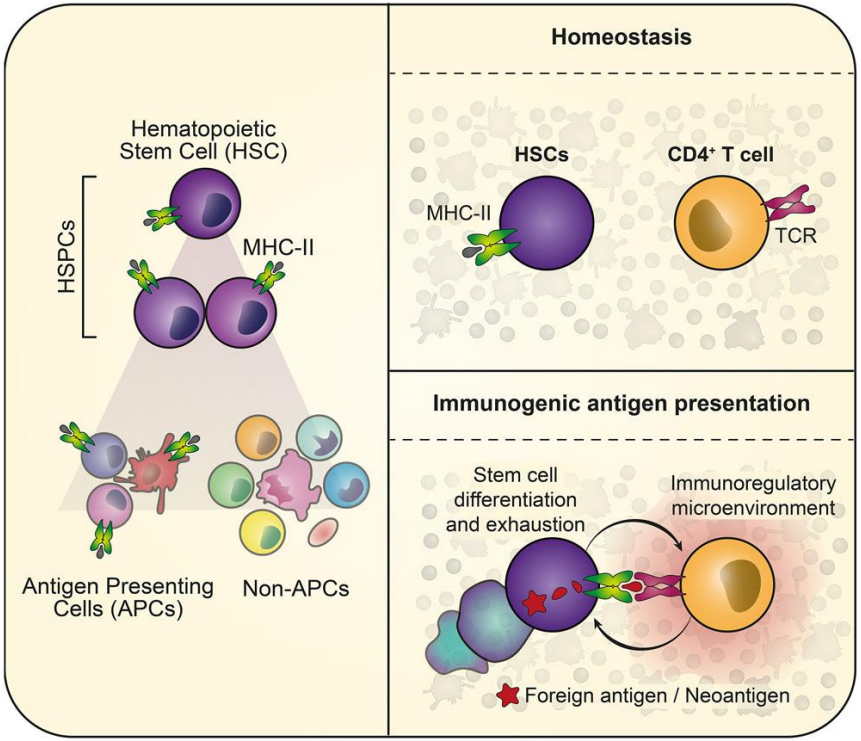
Rutella S et al, J Clin Invest, sept 13 2022



Tregs in AML: is it time for immunomodulation?

Although the notion that Tregs immunosuppression represents a crucial point in AML immune microenvironment, the mechanisms underlying Tregs induction are still poorly elucidated and largely unknown.

Ustun C et al. Blood 2011;118:5084-5095



Antigen presentation safeguards the integrity of the hematopoietic stem cell pool

HSPCs constitutively present antigens via MHC-II

Presentation of immunogenic antigens results in the activation of $CD4^+$ T cells

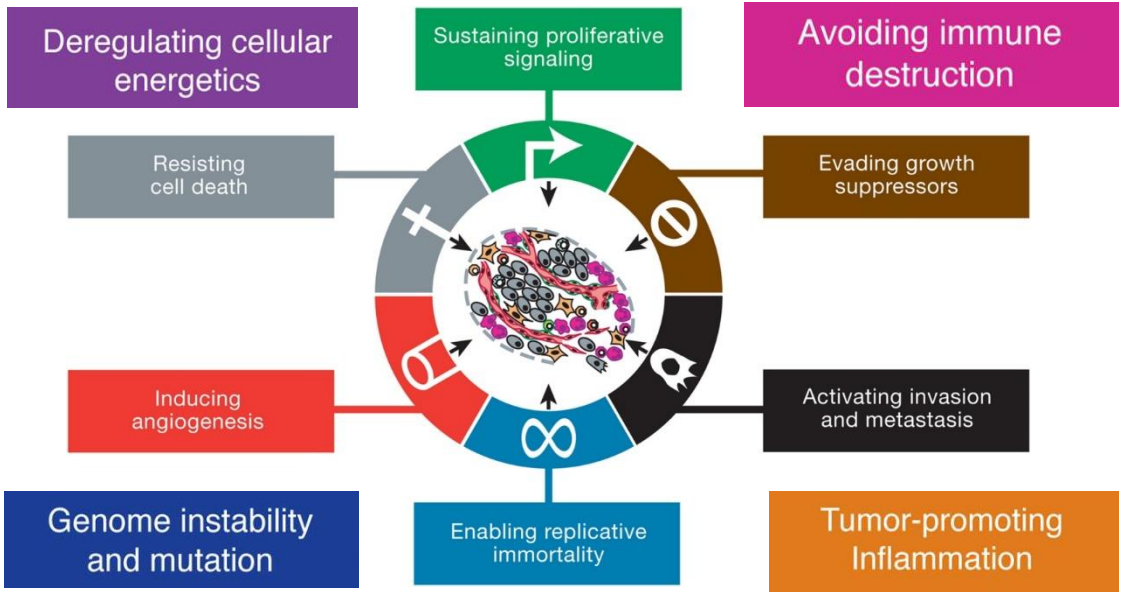
Antigen presentation causes differentiation and depletion of immunogenic HSPCs

This prohibits the onset of HSC-derived leukemias presenting neoantigens via MHC-II

CD4+ T cells activated by HSPCs confirmed that they acquired an immunoregulatory and anti-inflammatory phenotype

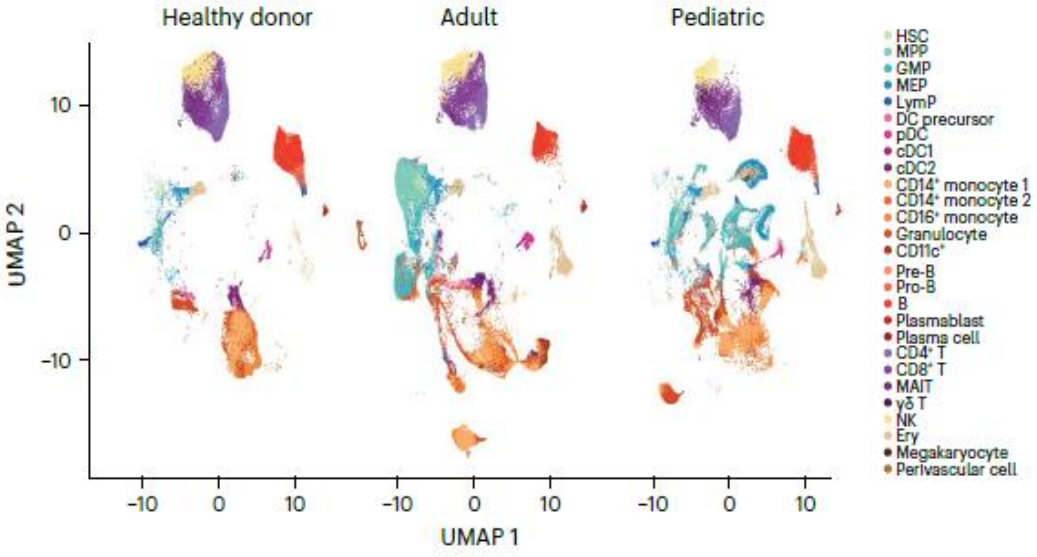
Hernandez-Malmierca et al, Cell Stem Cell, 29, 2022, Pages 760-775

Emerging Hallmarks of Cancer



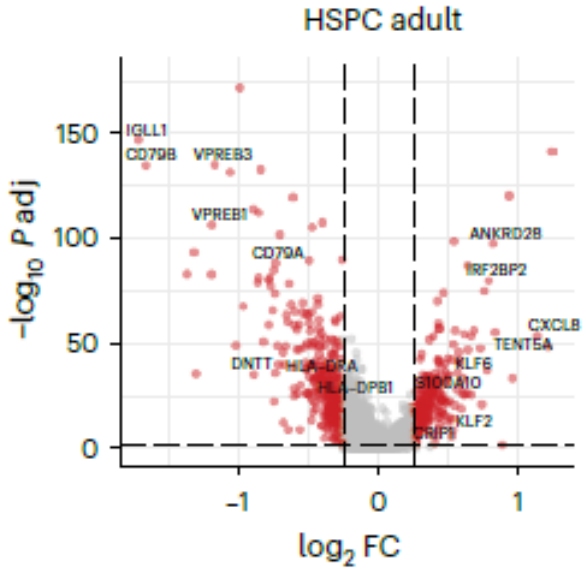
Cell 2011 144, 646-674

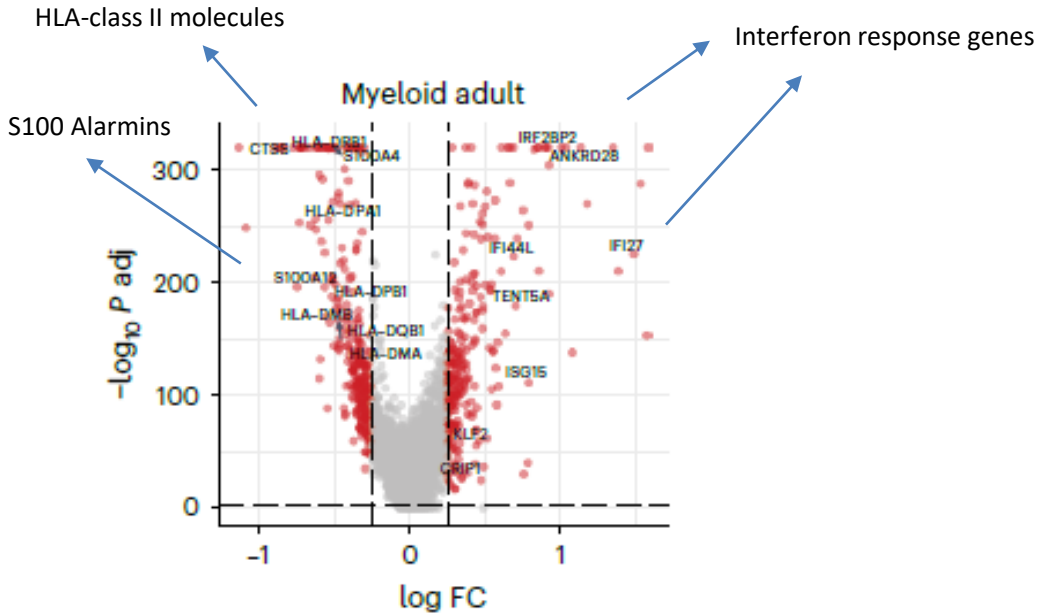
BM immune microenvironment is strongly altered in patients with AML by using a scRNA-seq approach



Lasry A et al, Nature Cancer, January 2023, 27-42

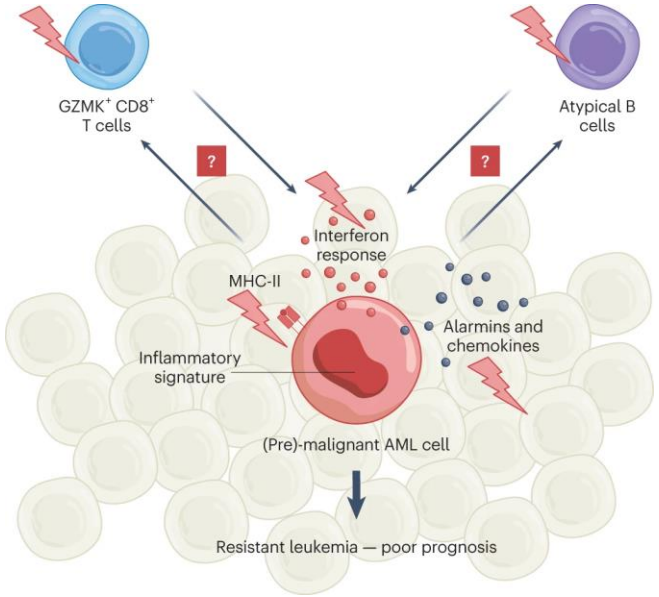
Dysregulated expression of genes associated with inflammatory pathways is a hallmark of AML





Lasry A et al, Nature Cancer, January 2023, 27-42

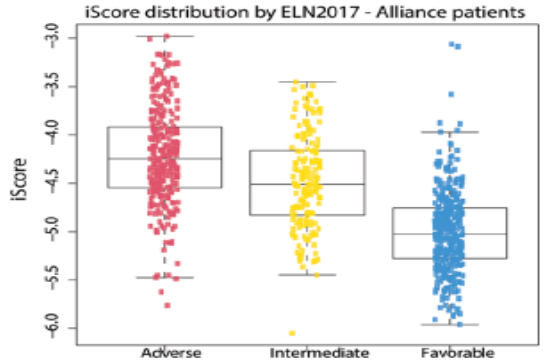
Atypical B cells and exhausted GZMK+ CD8 T cells are expanded in highly inflamed AML microenvironment



Although the relationship between the intrinsic inflammatory signature and the immune microenvironment is not well characterized, a complex interaction with bi-directional impact (arrows) likely exists, which results in resistant leukemia.

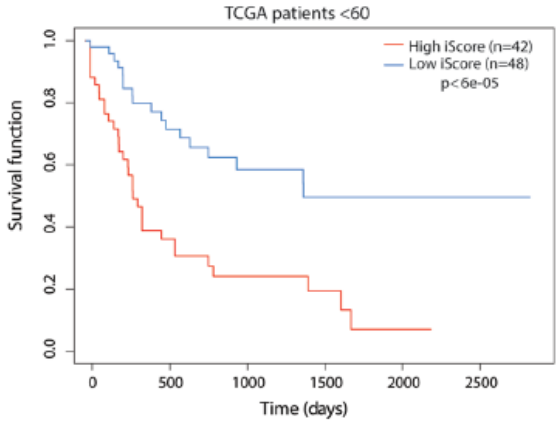
These discoveries pave the road for potential medical interventions, either directly on the intrinsic inflammatory pathways of the leukemic cell, or by targeting the immune cells.

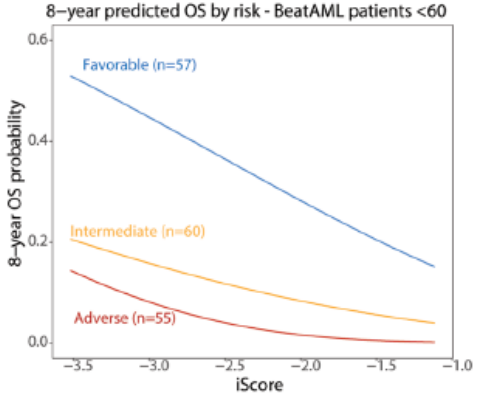
Asaf D. Yanir & Shai Izraeli Nature Cancer, 2023, pages 3–4

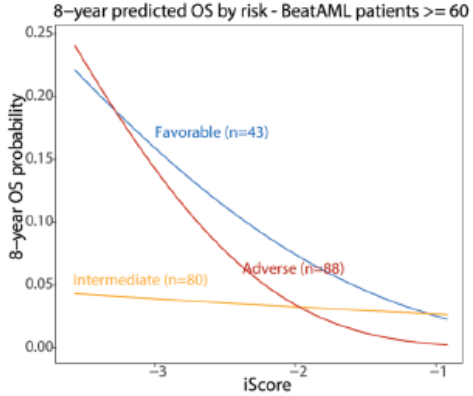


High inflammatory score is associated with adverse ELN risk group

High inflammatory score prognostically stratifies AML patients

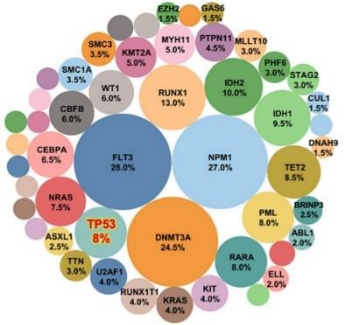


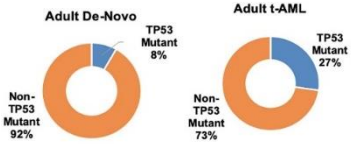


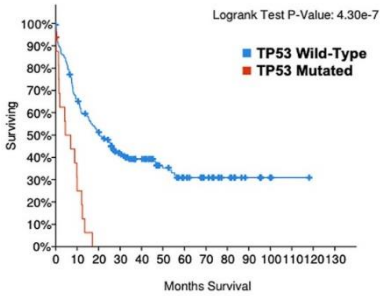


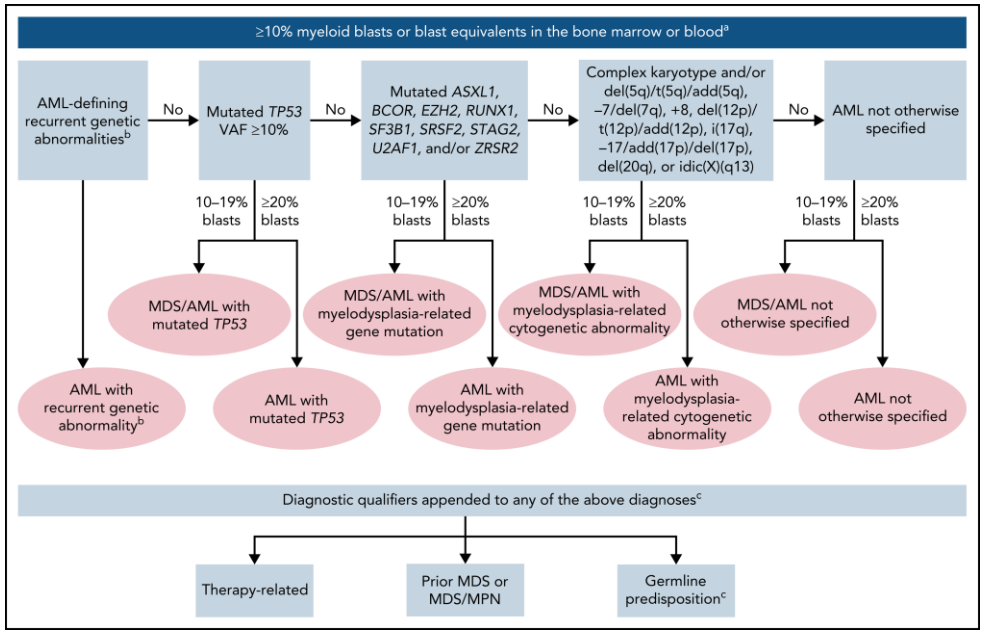
Lasry A et al, Nature Cancer, January 2023, 27-42

AML with mutated *TP53:* a separate entity in ICC AML classification



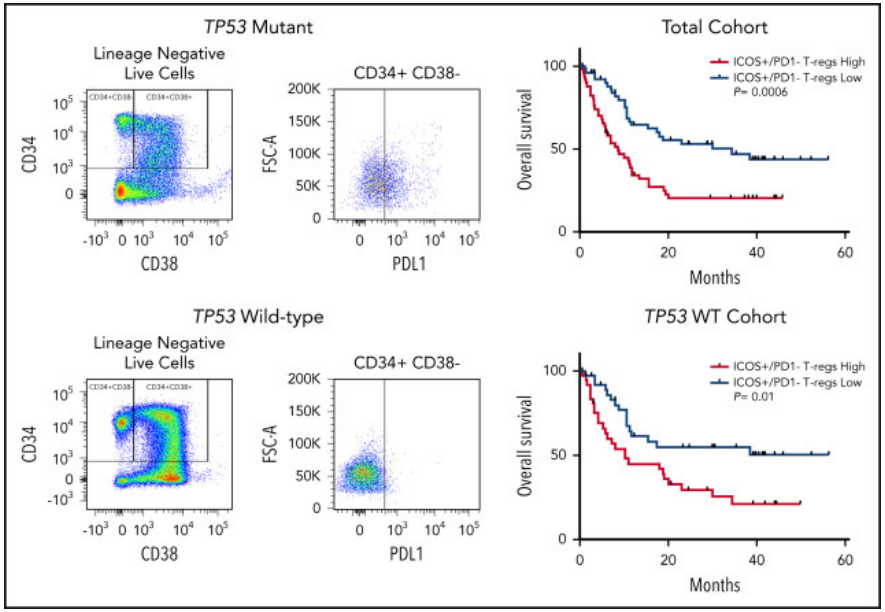


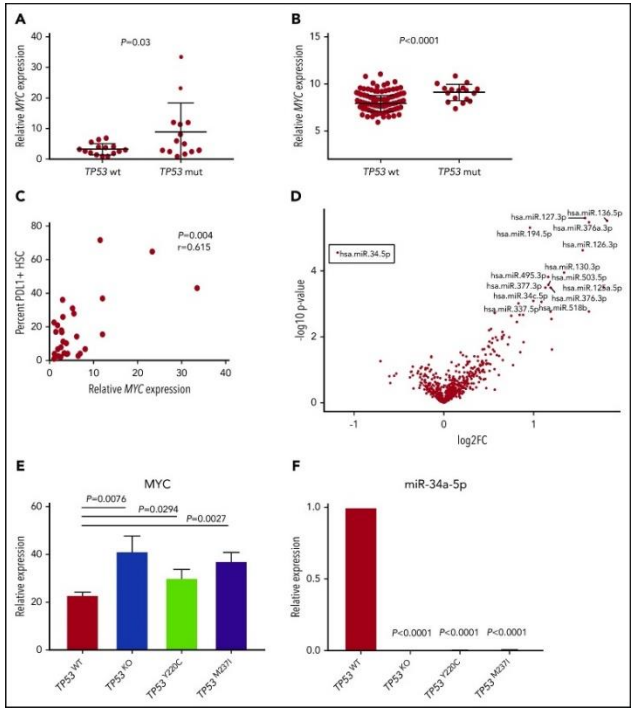




Arber DA et al, Blood. 2022;140(11):1200-1228.

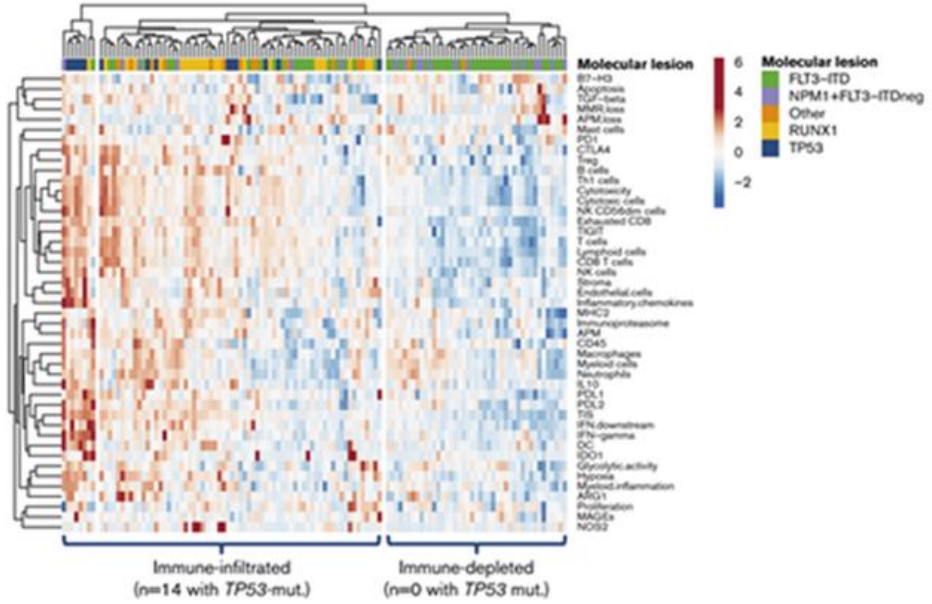
TP53 mutations in myelodysplastic syndromes and secondary AML confer an immunosuppressive phenotype

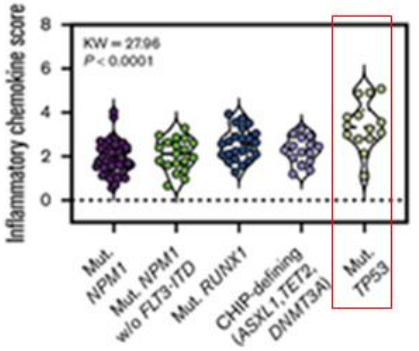


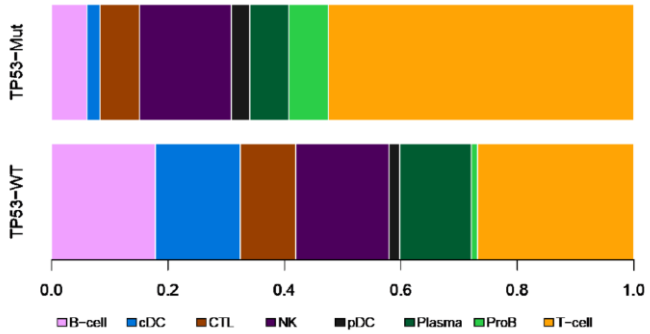


Sallman D et al, Blood. 2020 Dec 10;136(24):2812-2823

TP53mut AML patients show an inflammatory immune microenvironment

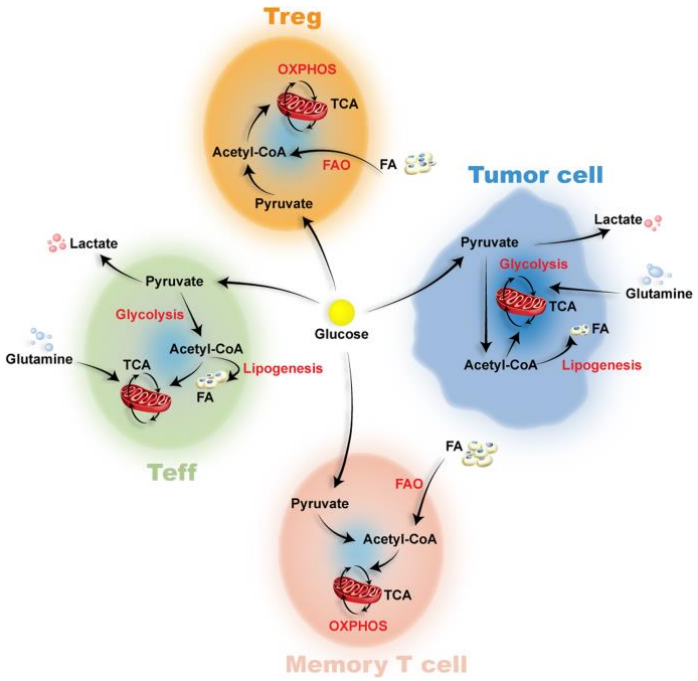




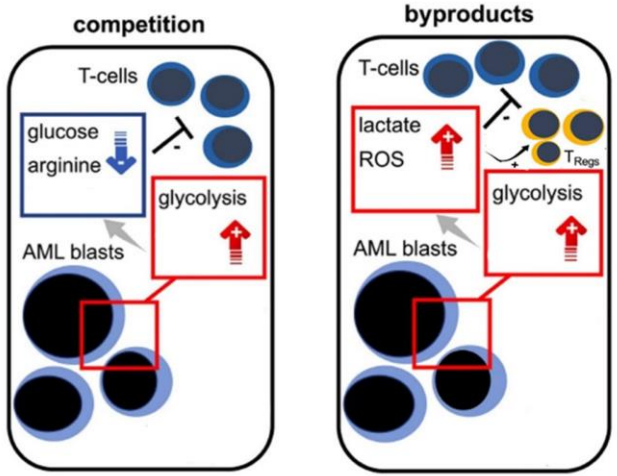


Vadakekolathu J et al. Blood Adv. 2020 Oct 27;4(20):5011-5024

AML metabolic alterations results in microenviromental metabolic remodelling in immune cells

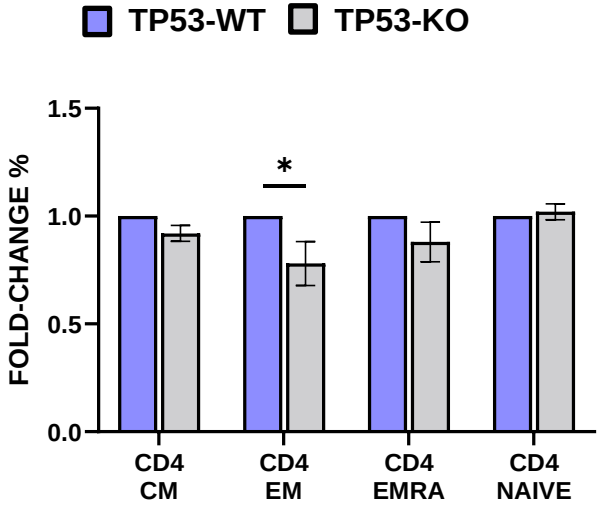


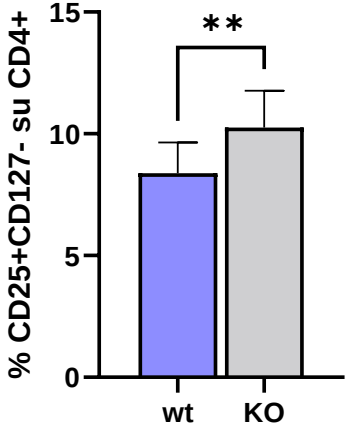
Which mechanisms?

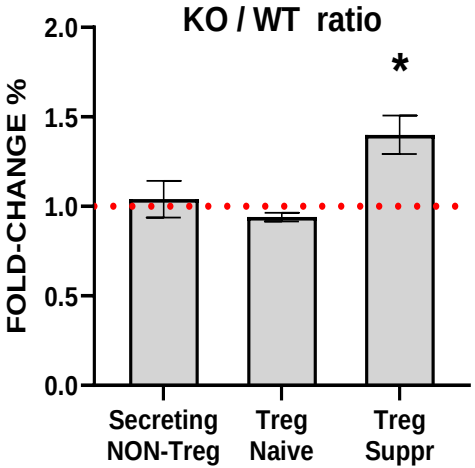


Mougiakakos D. Front Oncol. 2019 Nov 6;9:1166.

OCI-AML3 TP53 KO reduces the frequency of effector T cells and drives T reg expansion

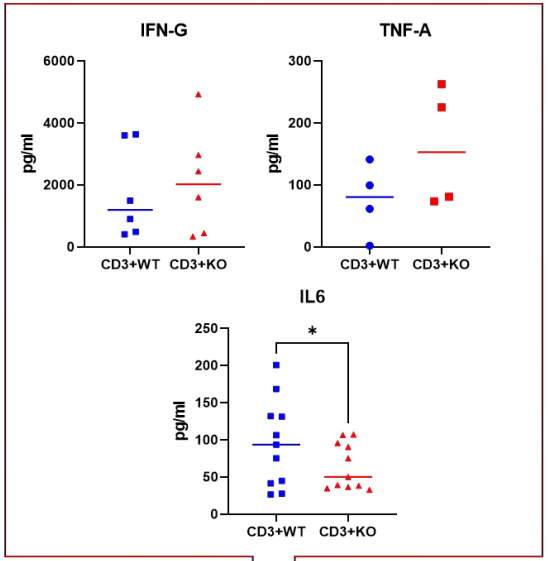


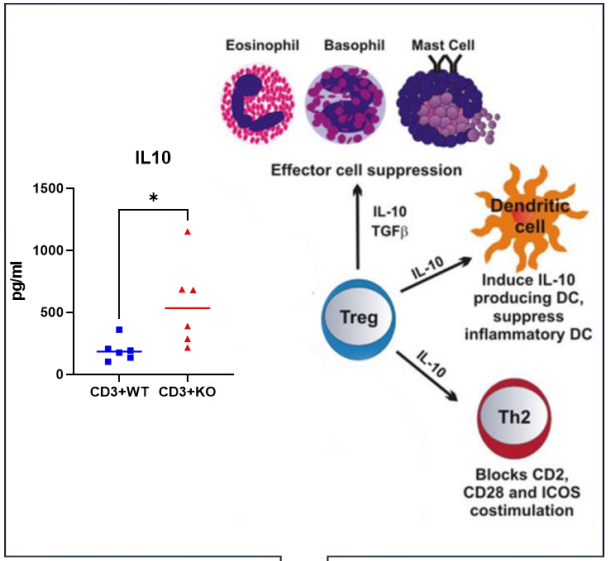




Salvestrini et al. In preparation

T cells co-cultured with OCI-AML3 TP53KO have increased secretion of inflammatory and tolerogenic cytokines



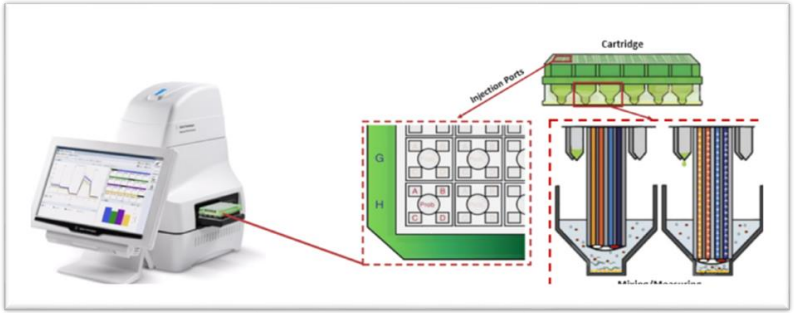


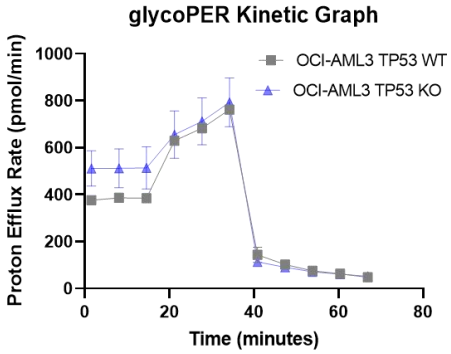
Inflammation

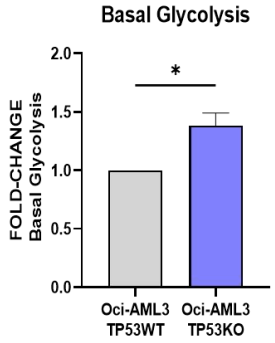
Tolerance

Salvestrini et al. In preparation

OCI-AML3 TP53KO cells have a preferential glycolytic metabolic profile

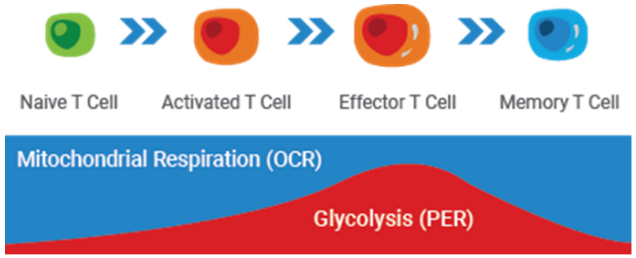


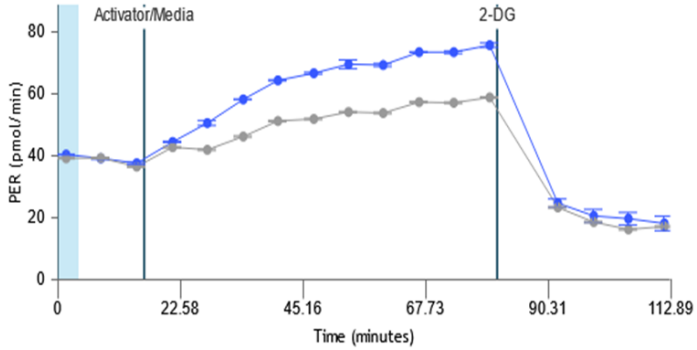


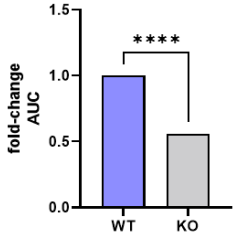


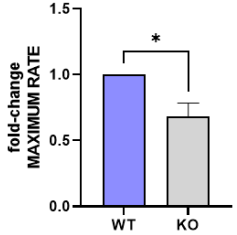
Salvestrini et al. In preparation

OCI-AML3 TP53 KO reduce the activatory potential of effector T cells



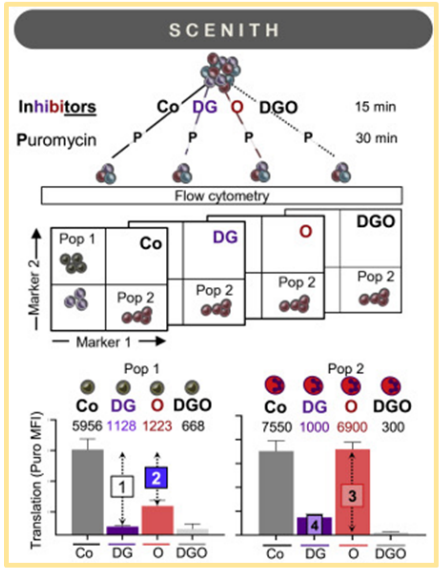


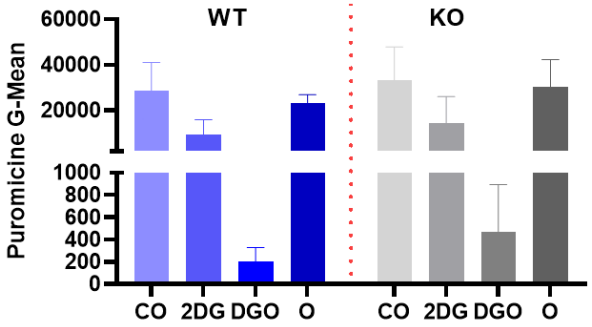


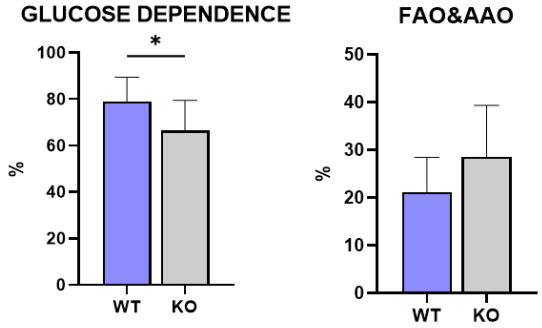


Salvestrini et al. In preparation

OCI-AML3 TP53KO reduce glucose dependence of Tregs which preferentially utilize FAO for bioenergetics



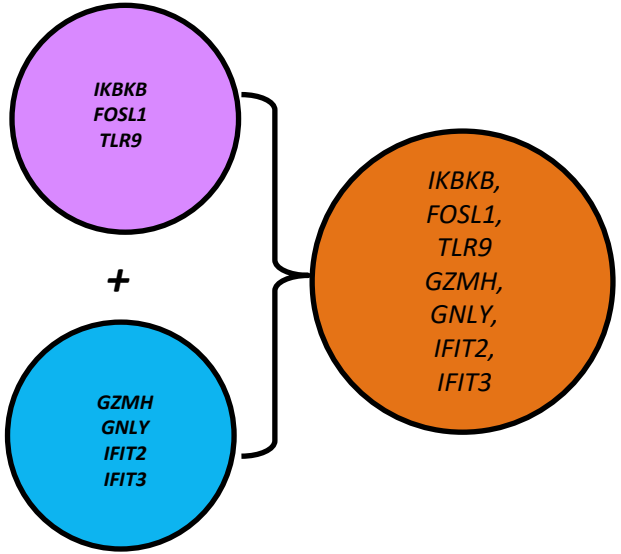




Salvestrini et al. In preparation

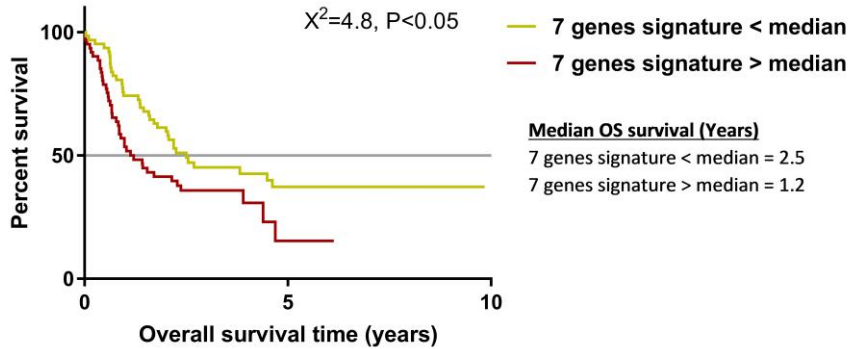
IDO1-PLXNC1 Nanostring® analysis identifies a 7-gene signature predicting outcome in AML

DE between PLXNC1^{high} and PLXNC1^{low}



DE between IDO1^{high} and IDO1^{low}

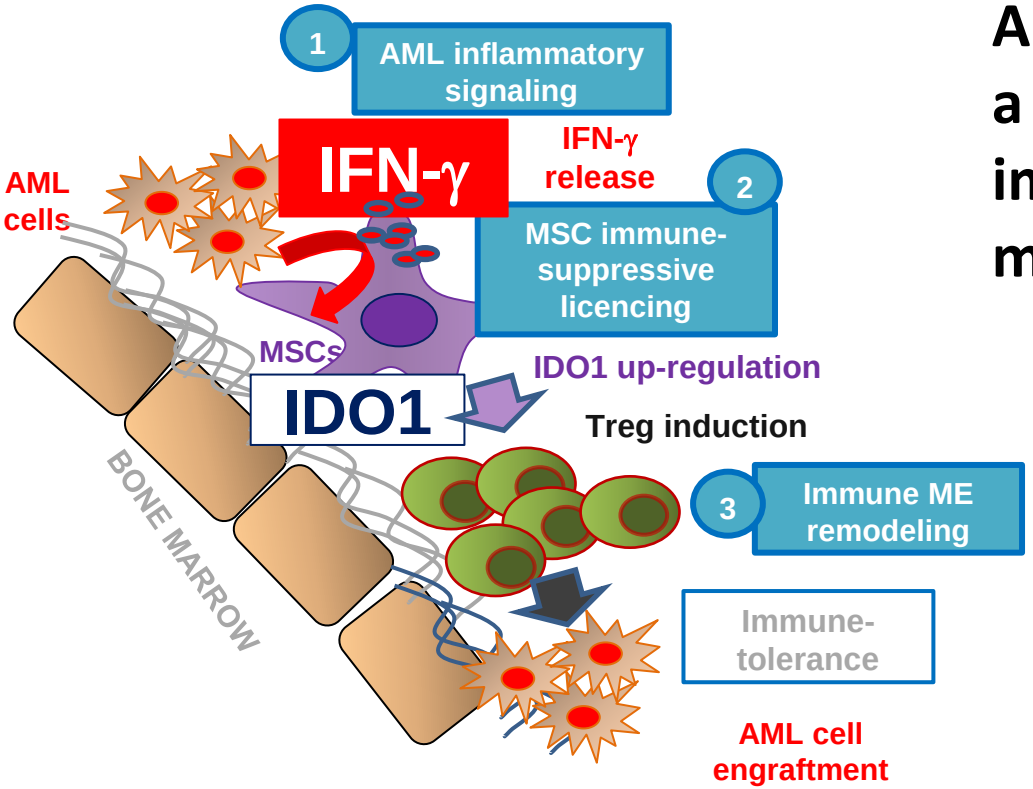
AML TCGA dataset



Numbers at risk

7 genes signature < median	62	12	0
7 genes signature > median	61	3	0

Ragaini et al, Blood Adv. 2022 Jan 11;6(1):87-99

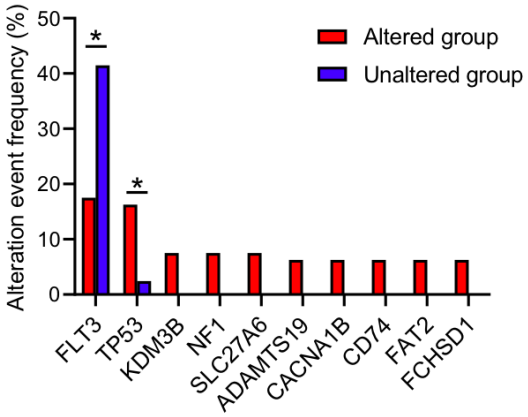


AML derived IFN-γ: a double-edged sword within immune BM microenvironment of AML

Along with activating pathways, IFN-γ-dependent signals produced by AML cells modify MSC functions and favor an immune-modulating milieu

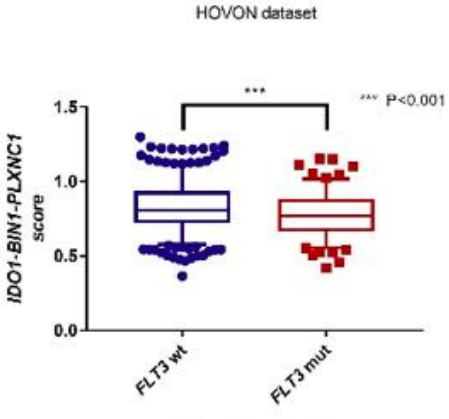
Corradi G et al. Clin Cancer Res, March 29, 2022

Correlation between DEGs in IFNG^{high} vs IFNG^{low} cases and *FLT3* mutational status (P*<0.001)**

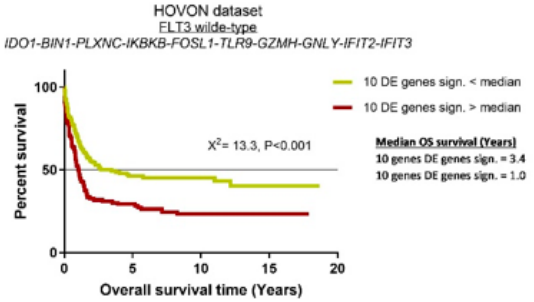


Corradi G et al. Clin Cancer Res, March 29, 2022

Score values significantly different according to FLT3 mutational status



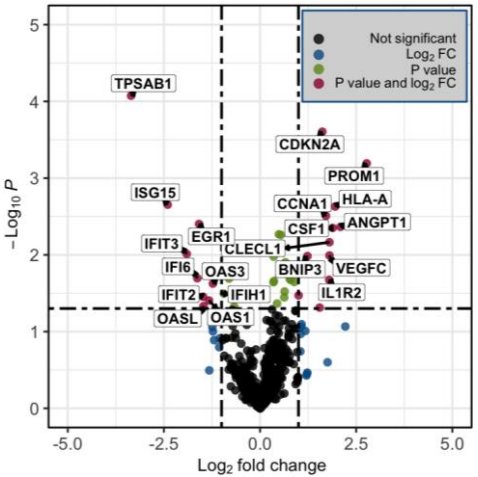
Among FLT3 wt: the score remained statistically significant



Ragaini et al, Blood Adv. 2022 Jan 11;6(1):87-99

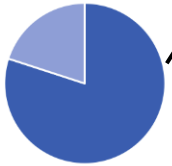
Response to Gilteritinib is associated with cell-extrinsic pathways involving innate immunity

***Volcano Plot from DEG analysis*:**
Resistant versus **Sensitive**
(log_2 FC cut-off = 1.0; *P* value cut-off = 0.05)

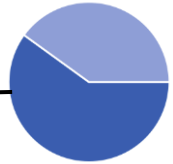


UP IN SENSITIVE **UP IN RESISTANT**

UP IN SENSITIVE



Interferon-related pathway (8/10)
Mostly $\alpha - \beta$
(ISG15, IFIT3, IFI6, OAS3, IFIT2, IFIH1, OASL, OAS1)

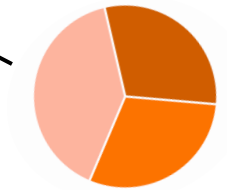


Innate Immunity
Viral response (6/10)
(TPSAB1, ISG15, OAS3, IFIT2, IFIH1, OAS1)

1 remaining tumor suppressor gene (EGR1)

UP IN RESISTANT

Oncogenes
(CSF1, VEGF, PROM1 and ANGPT1)

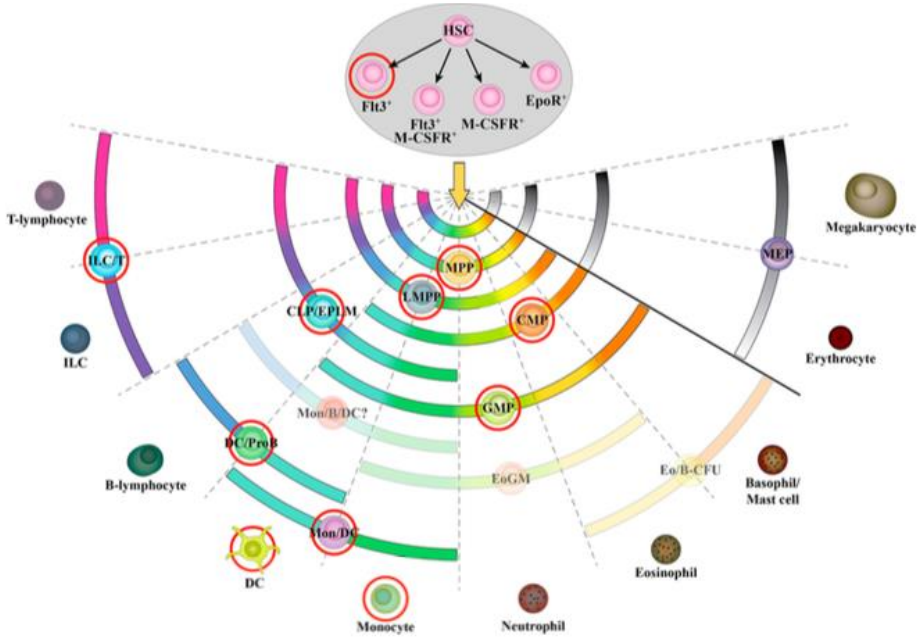


Cell cycle and apoptosis regulators
(CDKN2A, CCNA1 and BNIP3)

Adaptive Immunity
(HLA-A, CLEC-1 and IL1R2)

Nanni et al. In preparation

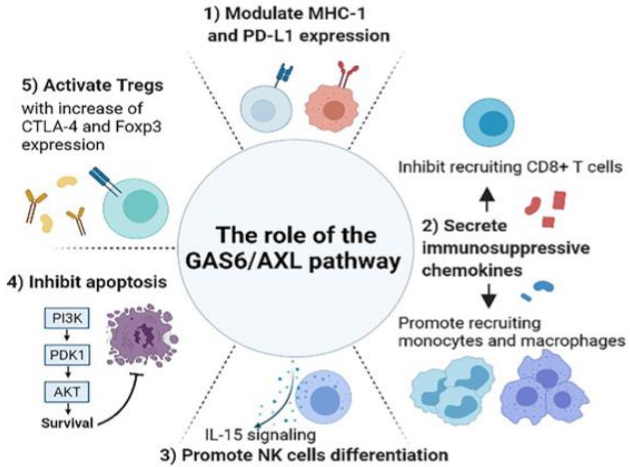
FLT3-FL signaling in normal hematopoieisis



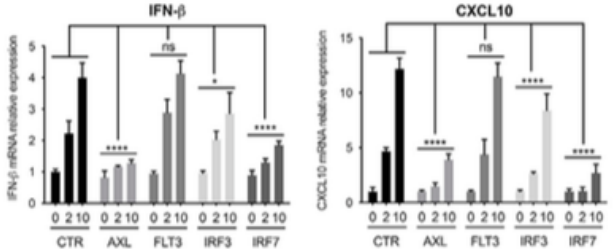
Panagiotis T. et al, Int. J. Mol. Sci. 2017, 18, 1115

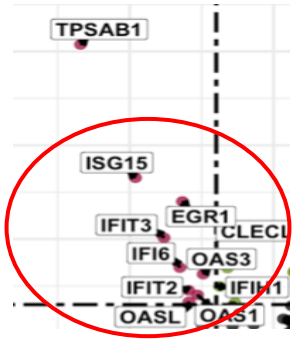
Gilteritinib is a selective and potent dual-inhibitor of FLT3 and AXL

AXL signaling exerts several immunomodulatory effects, with a prominent inhibition of innate immune response Chenjing Zhu et al, Molecular Cancer (2019) 18:153 Hye-Youn Son et al, Frontiers in Oncology, 11:756225.



Screening for enhancers of immune signaling by SARS-CoV-2: Gilteritinib the most potent stimulator, through the AXL-IRF7 axis.





In FLT3-mut AMLs sensitive to Gilteritinib we found upregulated the same Interferon-related genes

AXL long-term inhibition increases PD-1 expression on T-cells

POTENTIAL DRUG COMBINATION ?

Conclusions

- Immune microenvironment is emerging as critical component of BM niche
- A better understanding of cellular interactions is critical
- Driver mutations can modulate pathways which results in remodelling of immune microenvironment
- The immunometabolic perspective is an interesting area of investigation
- Therapeutic strategies should consider the impact of new and old drugs on immune microenvironment

Seragnoli Institute (Director: Prof. M. Cavo)
Acute Leukemia and MDS Group

Laboratory
Valentina Salvestrini
Marilena Ciciarello
Darina Ocadlikova
Dorian Forte
Giulia Rosaci

Clinical Team
Cristina Papayannidis
Stefania Paolini
Sarah Parisi
Chiara Sartor
Gianluca Cristiano
Jacopo Nanni
Letizia Zannoni
Federico Zingarelli

Study coordinators
Cinzia Bonajuto
Claudia Romano
Francesco Ingletto

Collaborators:
Roberto Maria Pellegrino (University of Perugia)

Francesco Fabbri
Giorgia Simonetti
Giovanni Martinelli
(IRCCS Meldola)

Sabina Sangaletti
Mario Colombo
(INT Milan)

Simón Mendéz-Ferrer (University of Cambridge)

Paolo Gallipoli
(Barts Cancer Institute, London)

Jayakumar Vadakekolathu, Sergio Rutella (Nottingham Trent University, UK)

Alessandro Gulino, Claudio Tripodo (University of Palermo)

Policlinico S.Orsola-Malpighi

SERVIZIO SANITARIO REGIONALE EMILIA-ROMAGNA
Azienda Ospedaliero - Universitaria di Bologna
IRCCS Istituto di ricovero e cura a carattere scientifico

ALMA MATER STUDIORUM UNIVERSITÀ DI BOLOGNA

Beat-Leukemia.org
Multi language resources for leukemia fighters.

SIE Società Italiana di Ematologia

AIL BOLOGNA
ASSOCIAZIONE ITALIANA CONTRO LE LEUCEMIE LINFOMI E MIELOMA SEZIONE DI BOLOGNA ONLUS

CANCER RESEARCH UK | CAMBRIDGE CENTRE

AIRC

NHS Blood and Transplant

ALMA IDEA Junior grant

FATRO

FABBRI 1905

AGB Associazione Giuseppe Bigi ONLUS

ASH/Bigi memorial award 2019