HIGHLIGHTS IN EMATOLOGIA

TREVISO
7-8 NOVEMBRE 2025

L’immunoterapia nelle Leucemie Mieloidi Acute: a che punto siamo?

Antonio Curti

IRCCS Azienda Ospedaliero-Universitaria di Bologna

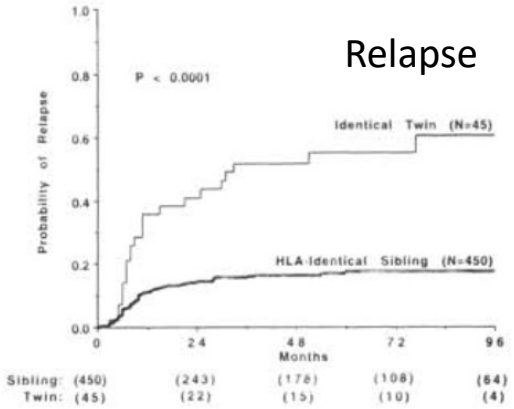
Istituto di Ematologia «Seràgnoli», Bologna

Disclosures of Antonio Curti

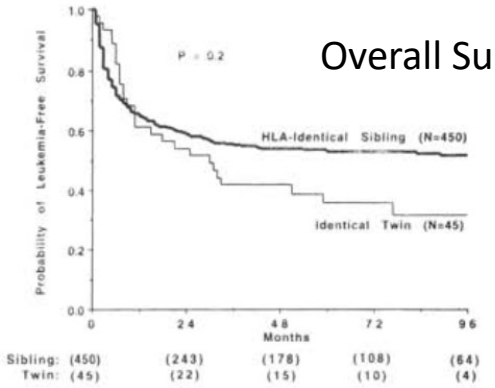
Company name	Research support	Employee	Consultant	Stockholder	Speakers bureau	Advisory board	Other
Abbvie	X					X	X
Jazz Pharma						X	X
Menarini-Stemline						X	X
Servier						X	X
Novartis							X
Pfizer	X					X	X

Immunosensitivity of AML: clinical evidence from allogeneic SCT

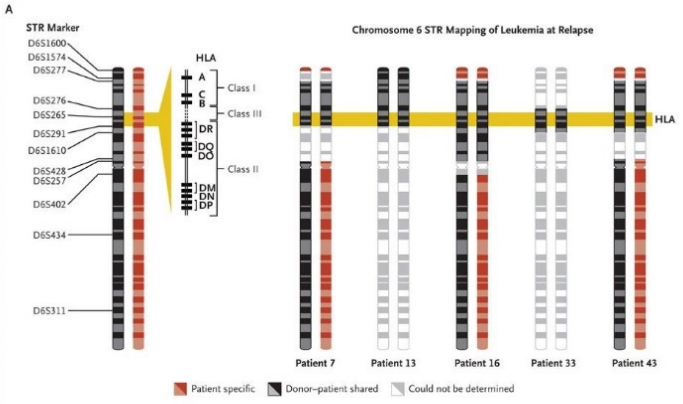
Relapse

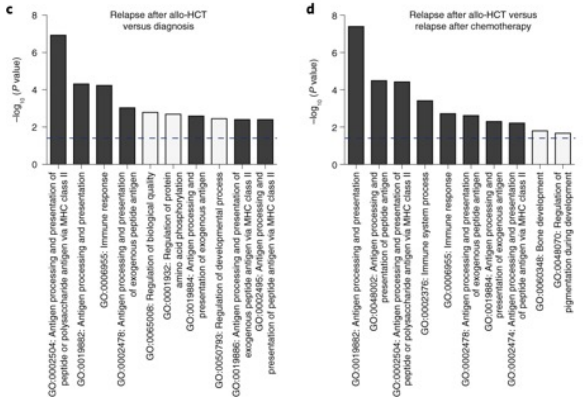


Overall Survival



RP Gale, 1994, Ann. Int. Med.





L. Vago, NEJM, 2009;
Toffalori, Nat. Med., 2019

Evolving immunological strategies to target AML cells

1) Immune checkpoint/macrophage blockade

2) Antigen-targeted immunotherapies
   -Leukemia vaccines
   -Bispecific or Dual-affinty T-cell engangers (BiTes/DARTS)
   -CAR T cells

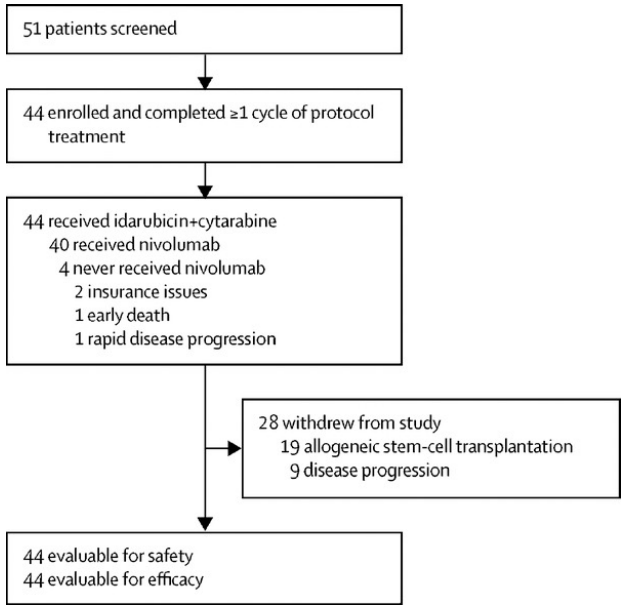
3) Inhibition of immunosuppressive factors

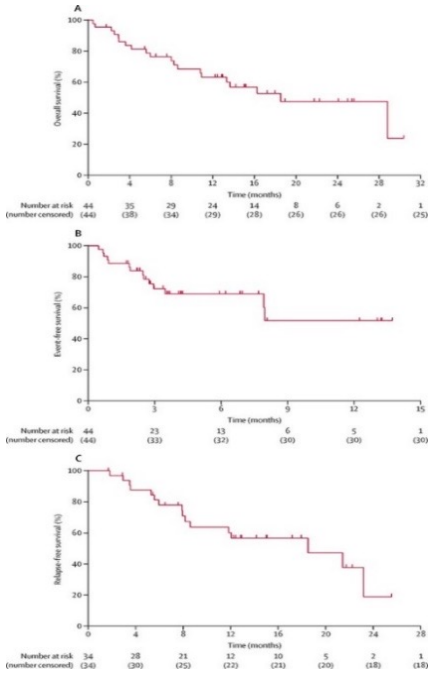
4) Cytokine therapies and adoptive transfer of NK cells

Pathways and targets for immunotherapies in AML

Pathway	Therapeutic action	Effects
PD-1/PD-L1/TIM-3/LAG-3	-mAb anti-PD-1/TIM-3/LAG-3 -mAb anti-PD-L1	- Increased T-cell cytotoxicity - Increased DC function as APCs
CD33	mAb anti-CD33	- AML cell lysis
CTLA-4	mAb anti-CTLA-4	- Increased T-cell cytotoxicity - Increased DC function as APCs
CD200	mAb anti-CD200	- Increased T/NK-cell cytotoxicity - Increased DC function as APCs
IDO	IDO1 inhibitor	- Prevention of T-cell tolerance
NK cells	adoptive cell therapy	- AML cell lysis
CAR-T cells	adoptive cell therapy	- AML cell lysis
Tregs	lymphodepletion therapy	- Prevention of T-cell tolerance
KIR	mAb anti-KIR	- AML cell lysis
Arginine	human recombinant arginase	- Prevention of immune tolerance
CIK cells	adoptive cell therapy	- AML cell lysis
TAAs (WT1, RHAMM..)	immunotherapy-peptide vaccines	- Specific AML cell lysis

Idarubicin, cytarabine, and nivolumab in patients with newly diagnosed AML or high-risk myelodysplastic syndrome: a single-arm, phase 2 study





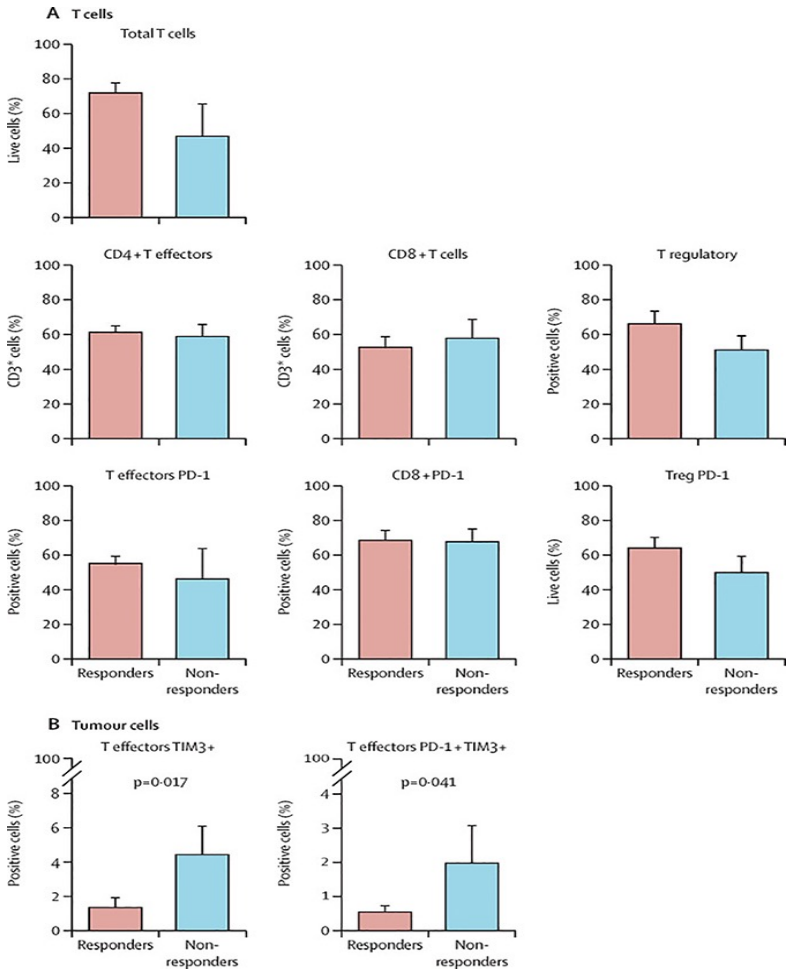
At a median follow-up of 17.25 months, median event-free survival was not reached.

The median relapse-free survival of responders was 18·54 months (1·7–25·6).

Highly favorable safety profile

19 responders of the 44 patients (43%) proceeded to allogeneic stem cell transplantation with grade 3/4 GvHD disease seen in 5.

Ravandi et al, Lancet Haematol. 2019 Sep; 6(9): e480–e488.

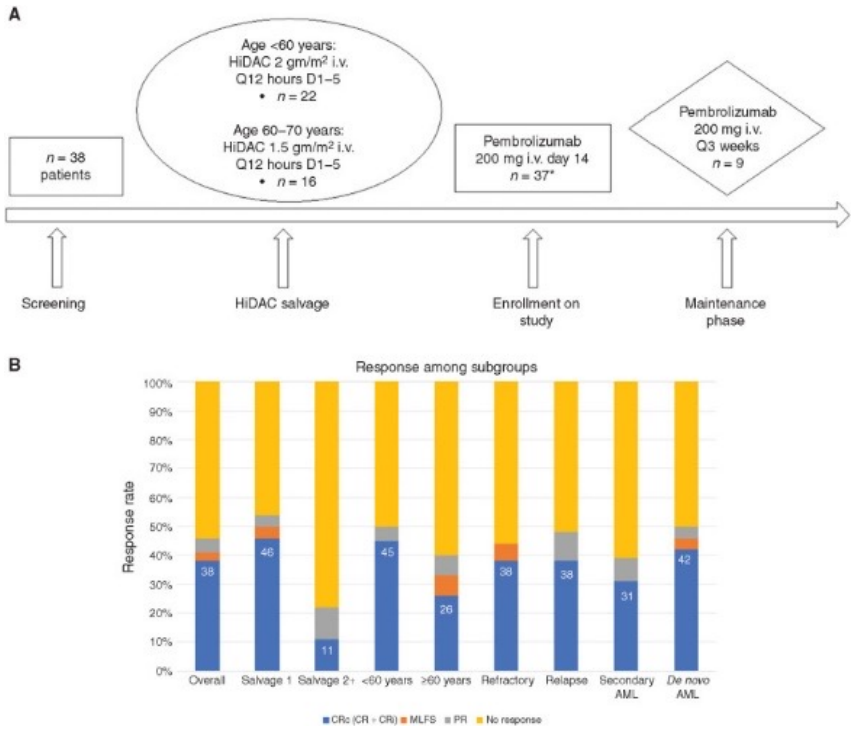


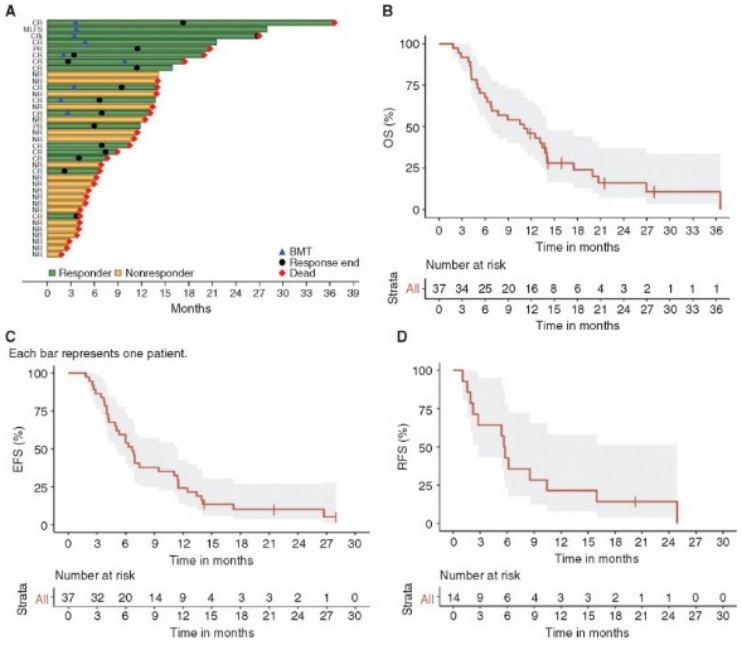
Idarubicin, cytarabine, and nivolumab in patients with newly diagnosed AML or high-risk myelodysplastic syndrome: a single-arm, phase 2 study

Non-responders had significantly higher percentage of CD4+ T-effectors co-expressing PD-1/TIM-3 (p=0·01) and PD-1/LAG-3 (p=0·04) compared to responders.

Ravandi et al, Lancet Haematol. 2019 Sep; 6(9): e480–e488.

Phase II Trial of Pembrolizumab after High-Dose Cytarabine in R/R AML

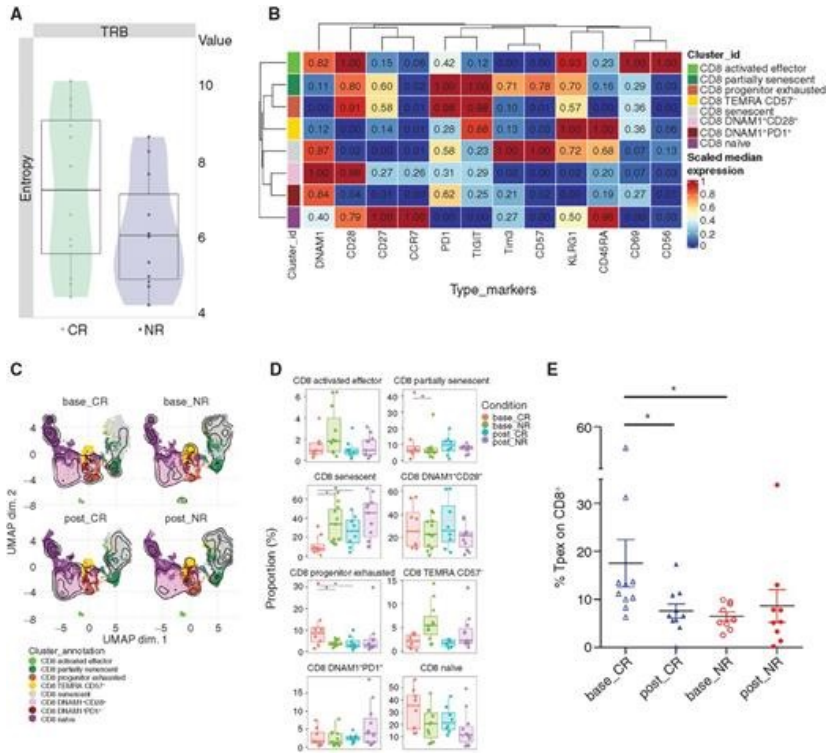




ORR and composite CR rates were 46% and 38%, respectively.
CRc rates were encouraging in those receiving treatment as first salvage therapy (13/28 = 46%), <60 years (10/22 = 45%), and refractory AML (6/16 = 38%).
In HiDAC-naïve patients, the CRc rate and median OS were 47% and 13.6 months, respectively

Zeidner et al, Blood Cancer Discov (2021) 2 (6): 616–629.

Phase II Trial of Pembrolizumab after High-Dose Cytarabine in R/R AML



TCR Vβ sequencing data revealed that patients who subsequently achieved CR had a trend toward higher TCR diversity at baseline compared with NR patients

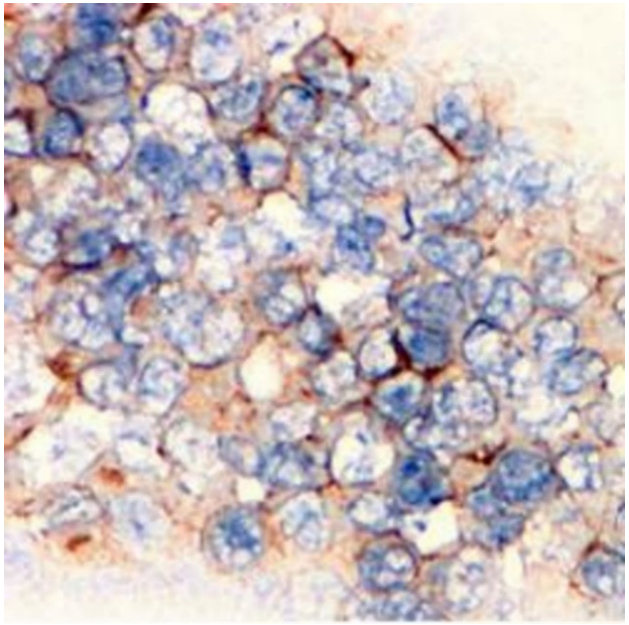
Patients who achieved CR had increased frequency of $CD8^+$ T cells expressing CD28, PD-1, and TIGIT, and lacking expression of Tim-3 and CD57 in the BM at baseline

$CD8^+CD45RA^-CD27^+$/int$CD28^+PD1^+TCF1^+$ T cells are increased in patients who achieved CR compared with NRs.

TCF-1 is a transcription factor essential for the stem-like properties of intratumoral $CD8^+$ T cells.
The presence of $CD8^+$ T cells coexpressing TCF-1 and PD-1 appears to be critical for immunotherapy response.

Zeidner et al, Blood Cancer Discov (2021) 2 (6): 616–629.

PD-L1 expression in MDS and AML cells is enhanced by HMAs

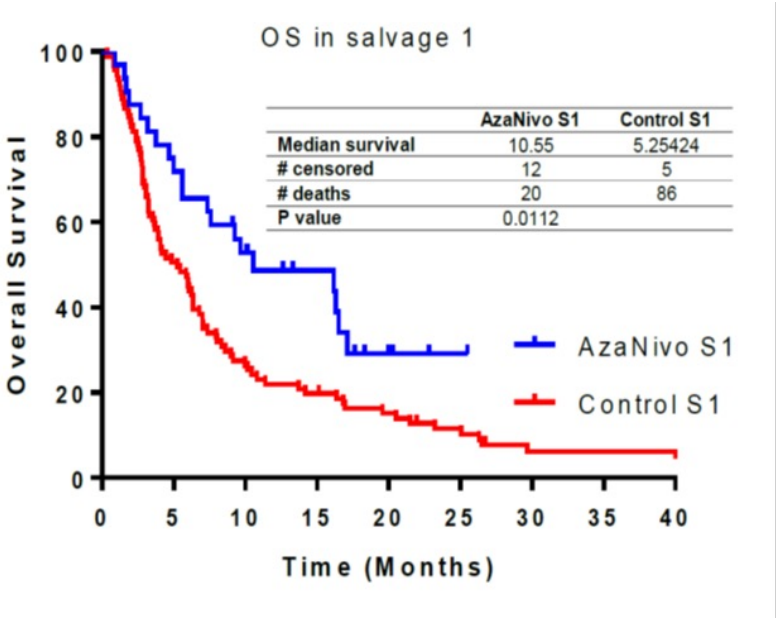


Carlos E. Bueso-Ramos et al. Blood 2013;122:2767

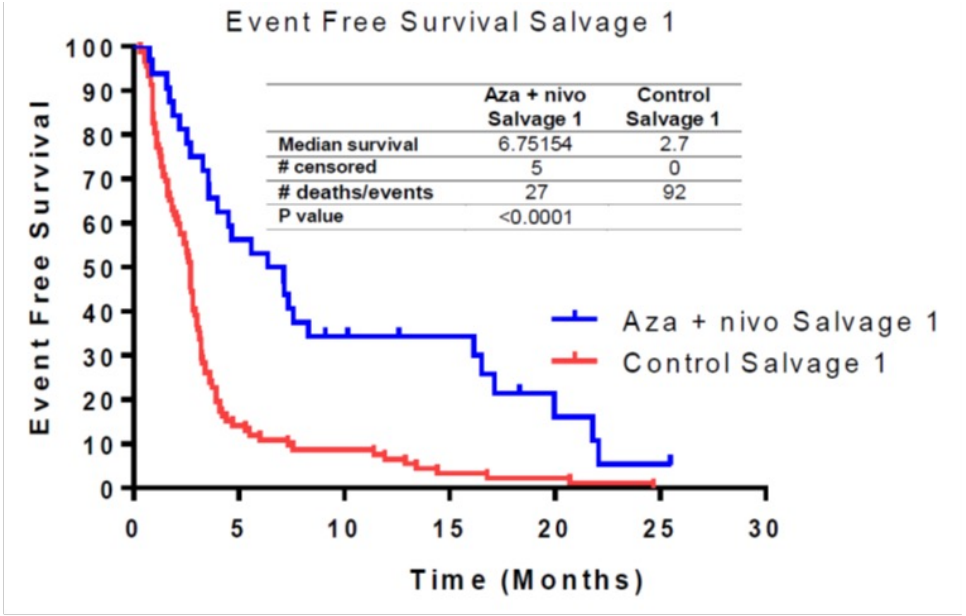
Exposure to decitabine resulted in demethylation of PD-L1 in AML cell lines, and the demethylation effect was also observed in HMAs treated MDS and AML patients

Expression of Immune Checkpoints PD-L1, PD-L2, PD-1 and CTLA4 Predict For Prognosis and Resistance To HAs In MDS

Efficacy, Safety, and Biomarkers of Response to Azacitidine and Nivolumab in Relapsed/Refractory AML: A Nonrandomized, Open-Label, Phase II Study



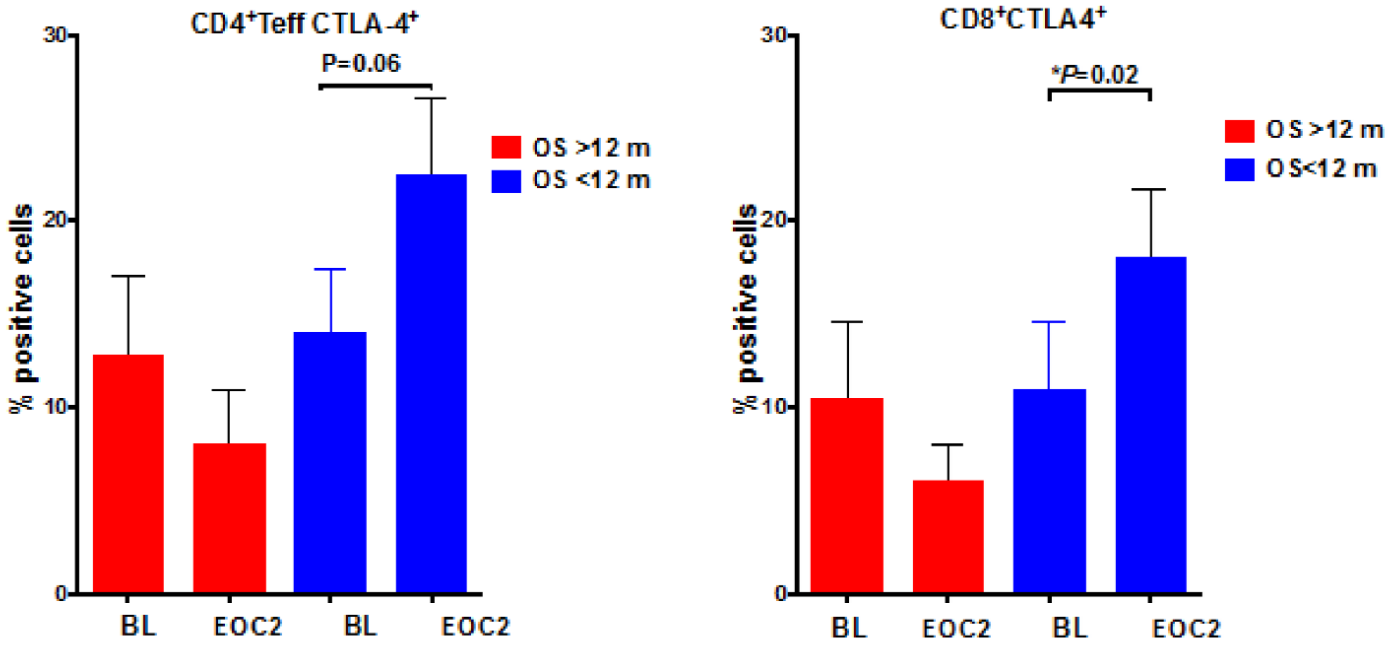
	AzaNivo S1	Control S1
Median survival	10.55	5.25424
# censored	12	5
# deaths	20	86
P value	0.0112	



	Aza + nivo Salvage 1	Control Salvage 1
Median survival	6.75154	2.7
# censored	5	0
# deaths/events	27	92
P value	<0.0001	

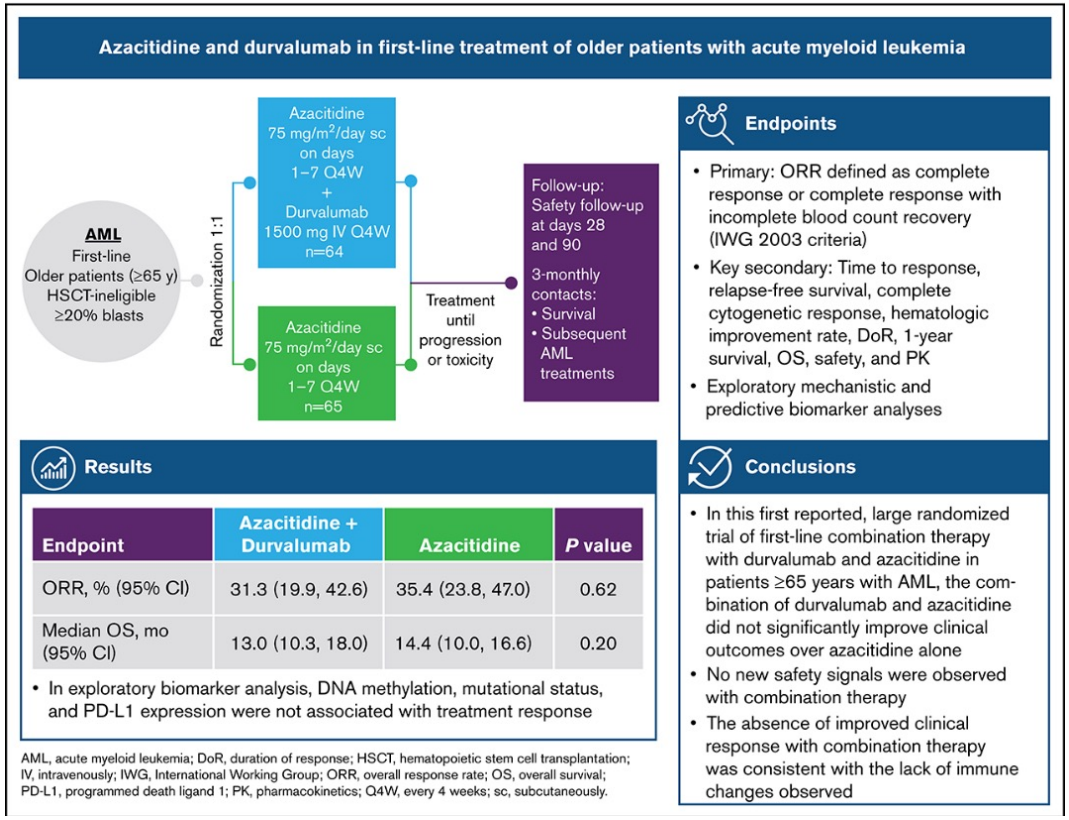
Daver N et al. Cancer Discov 2019;9:370-383

Efficacy, Safety, and Biomarkers of Response to Azacitidine and anti-PD1 in Relapsed/Refractory AML: a Nonrandomized, Open-Label, Phase II Study



Daver N et al. Cancer Discov 2019;9:370-383

A randomized phase 2 trial of azacitidine with or without durvalumab as first line therapy for older patients with AML

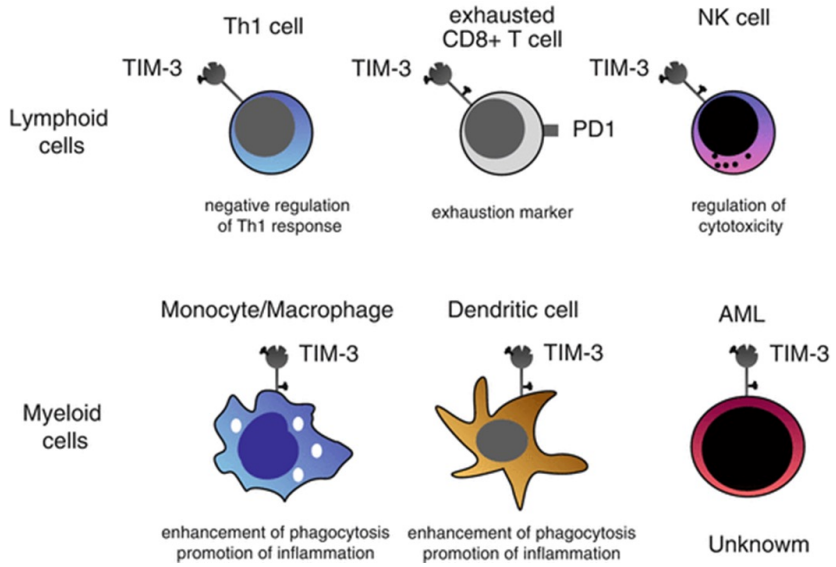


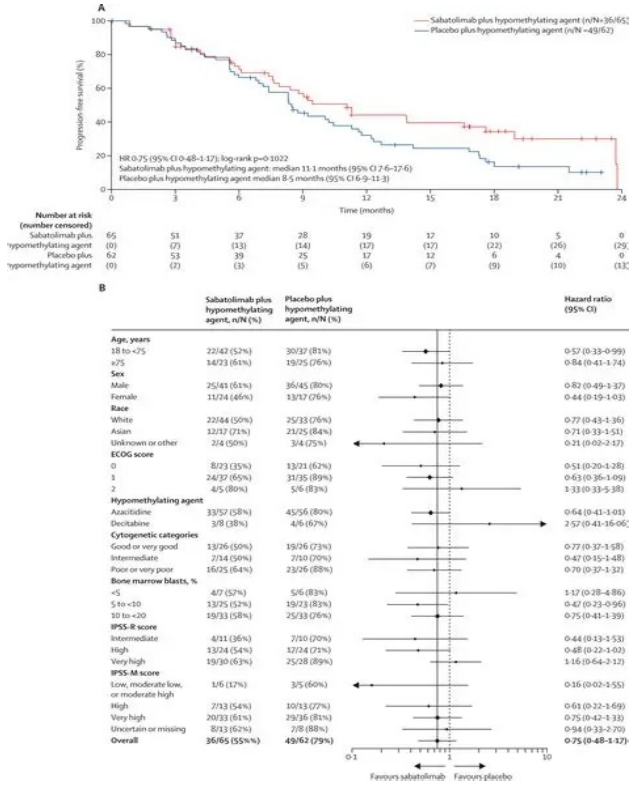
Key Points

- This is the first reported randomized trial of immune checkpoint inhibitor therapy in older patients with AML.
- Azacitidine combined with the PD-L1 inhibitor durvalumab was feasible but did not improve outcomes over azacitidine alone.

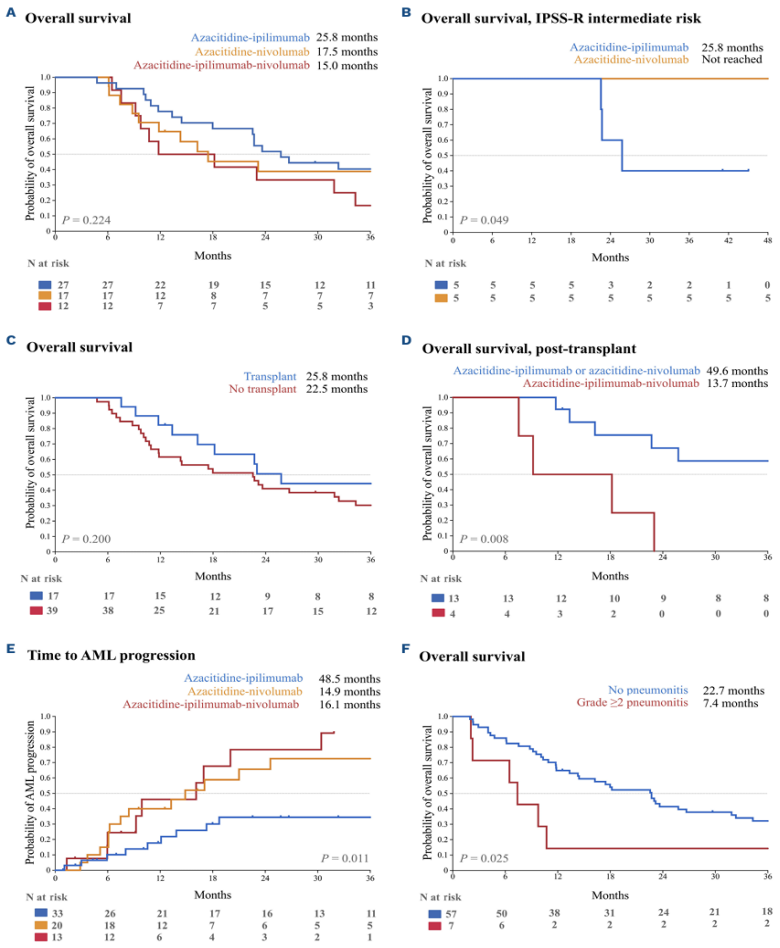
Zeidan AM et al, Blood Adv (2022) 6 (7): 2219–2229.

Sabatolimab plus hypomethylating agents in previously untreated patients with higher-risk myelodysplastic syndromes (STIMULUS-MDS1): a randomised, phase 2 trial





Zeidan A et al, Lancet Hematol, Volume 11, Issue 1e38, 2024



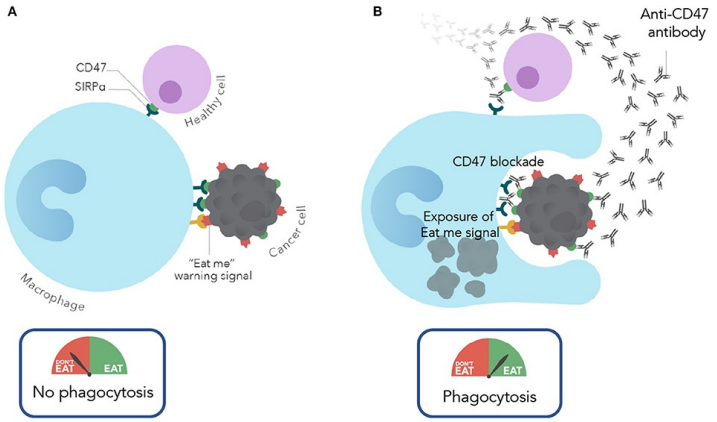
A phase II trial of azacitidine with ipilimumab, nivolumab, or ipilimumab and nivolumab in previously untreated myelodysplastic syndrome

- Azacitidine and nivolumab produced higher rates of CR and was associated with a greater survival benefit for IPSSR intermediate risk MDS than azacitidine ipilimumab.

- High grade toxicities was associated tothe triplet cohort than in the doublet cohorts, and the triplet regimen appeared to be associated with increased post transplant mortality

Bouligny IM et al, Haematologica. 2025 Jul 1;110(7):1628-1633

Phase II study of azacitidine, venetoclax and magrolimab for newly diagnosed and relapsed/refractory AML



Magrolimab (Hu5F9-G4) is an antibody blocking CD47, a macrophage immune checkpoint and "don't eat me" signal on cancers
Magrolimab induces tumor phagocytosis and eliminates LSCs. Azacitidine (AZA) synergizes with magrolimab by inducing "eat me" signals on leukemic blasts, thereby enhancing phagocytosis.

Chao MP et al, Front. Oncol., 22 January 2020

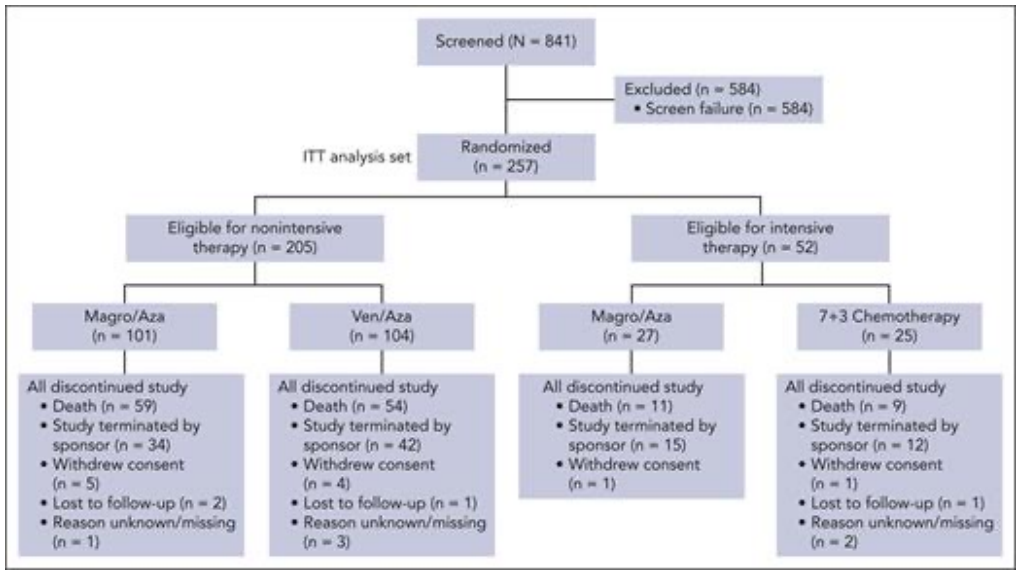
43 ND and 36 R/R patients

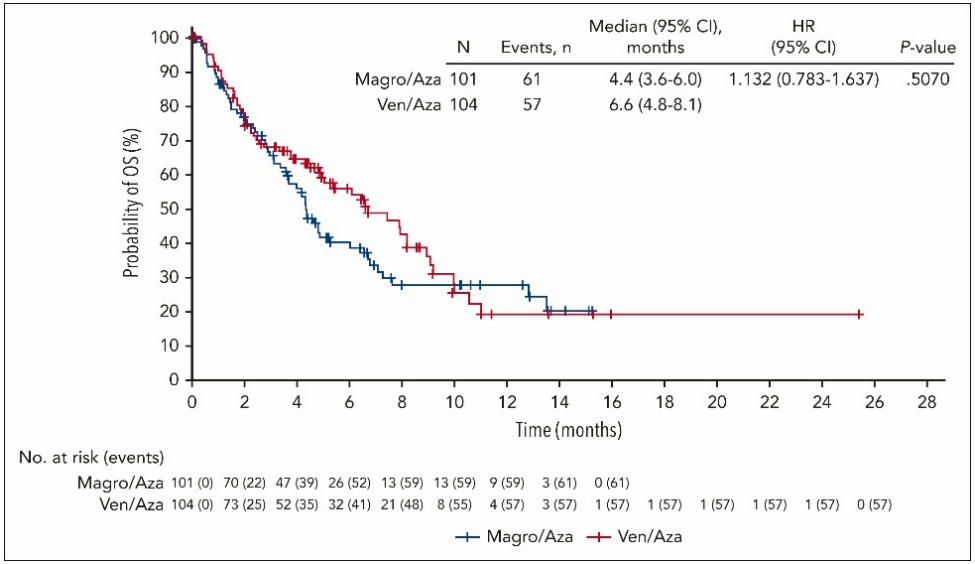
De novo AML: CR rate was 46% in patients with mutated *TP53* and 55% in those with wild-type *TP53*. Among patients with secondary AML, the CR rates were 40% and 60%, respectively.
The median duration of response was not reached regardless of *TP53* status. The 12-month overall survival (OS) rate was 83% in patients with wild-type *TP53* and 53% in *TP53* mut.

R/R AML: 18 patients were previously exposed to venetoclax and 18 were venetoclax-naïve. The CR rate was 19% overall, 0% in the venetoclax-exposed cohort, and 39% in the venetoclax-naïve. The median relapse-free survival was 3.1 months in the exposed cohort and 7.5 months in the naïve cohort. The median OS was 3.1 months and 5.6 months, respectively.

Daver N, et al, ASH 2022

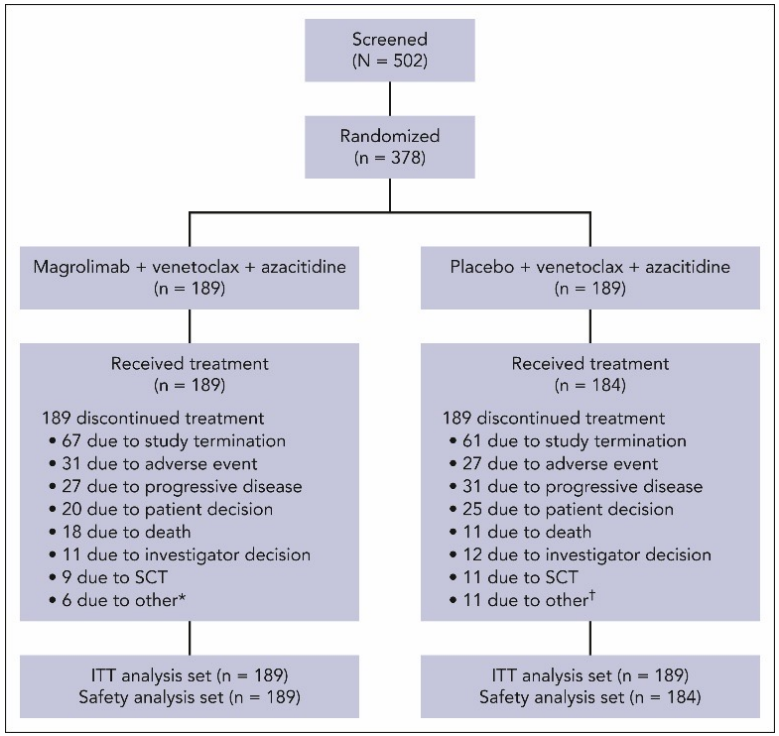
Magrolimab plus azacitidine vs physician's choice for untreated TP53-mutated acute myeloid leukemia: the ENHANCE-2 study

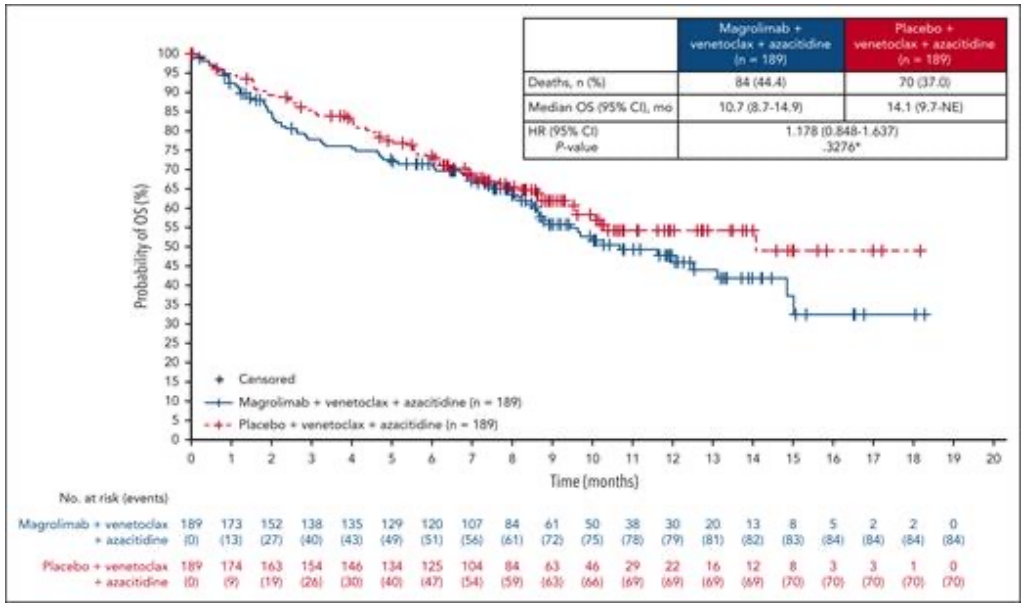




Zeidner JF et al, Blood (2025) 146 (5): 590–600.

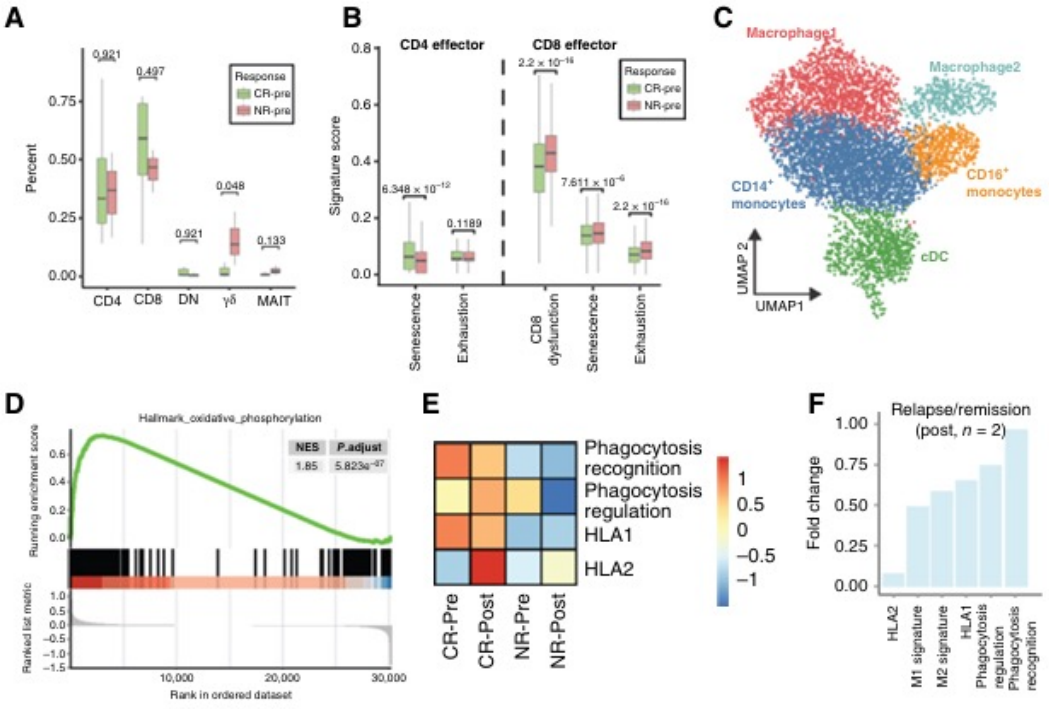
The ENHANCE-3 study: venetoclax and azacitidine plus magrolimab or placebo for untreated AML unfit for intensive therapy





Daver N et al, Blood (2025) 146 (5): 601–611.

Extrinsic factors in the tumor microenvironment affect patients' response to azacitidine, venetoclax and mabrolimab



Features associated with response:

T-cells: low exhausted and senescence signatures in CD8 effector cells (role of T cells in response)

Nonmalignant myeloid cells

- Oxidative phosphorylation phenotype
- Phagocytosis recognition
- HLA1 and HLA2 expression (antigen presentation)

Clin Cancer Res; 31(12) June 15, 2025

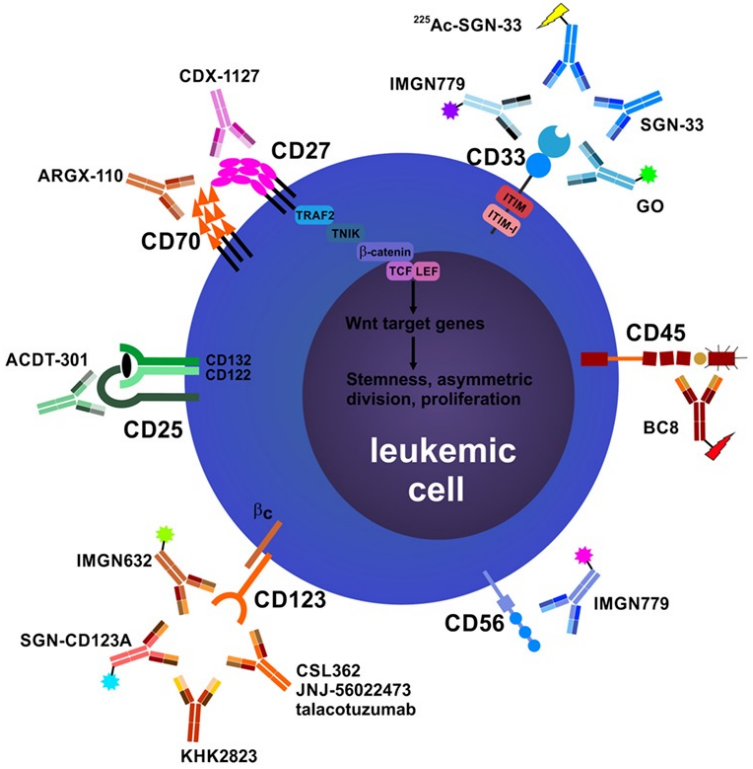
T cell–redirecting therapies in AML: T-cell engagers

Ab type	CD33			CD123			CD123	CLL-1
	AMG330[1]	AMG 673[2]	AMV-564[3]	Flotetuzumab[4]	JNJ-63709178[5]	Vibecotamab[6]	SAR443579[7-9]	MCLA-117[10]
Structure								
Manufacturer	Amgen	Amgen	Amphivena	Macrogenics	Janssen	Xencor	Innate/Sanofi	Merus
Phase	1	1	1	1, RP2D	1	1/2	I/II	1
N	96	46	53	246	62	106	23	62
Histology	r/r AML, MRD⁺ AML	r/r AML	r/r AML	r/r AML	r/r AML	r/r AML, B-ALL, CML	r/rAML, B-ALL and MDS	r/r AML, ND elderly
Prior Therapies	≥1	≥4	≥1	≥2	1-10	1-8	1-10	0-≥4
CRS (grade ≥3)	67% (13%)	50% (13%)	n.a. (0%)	50% (7%)	44% (15%)	58% (15%)	9% (n.r.), IRR 43%	36% (9%)
ORR	**19%**	**44% (12/27)**	**49%**	**30%**	**n.a.**	**>0.75 µg/kg 14% (7/51)**	5% (0 %)	**n.a.**
CR/CR$_i$	**17% (7/42)**	**4% (1/27)**	**6% (2/35)**	**27% (8/30)**	**0% (0/62)**	**10% (5/51)**	**12 % (3/23)**	**0% (0/58)**

1. Ravandi, Subklewe et al, Stein; Leukemia and Lymphoma 2024. 2. Subklewe M, et al. ASH 2019. Abstract #833. 3. Westervelt P, et al. ASH 2019. Abstract #834. 4. Uy GL, et al. Blood 2021. 5. Boyiadzis M, et al. Clin Transl Sci. 2023. 6. Ravandi F, et al. ASH 2020. Abstract #460. 7. Labrijn et al, Nature Reviews Drug Discovery 2019. 8. Stein et al, ASH 2024. Abstract #2883. 9. Bajel et al, ASH 2023 Abstract #3474. 10. Mascarenhas J, et al. EHA 2020. Abstract #538;

Subklewe M … Curti A, Semin Hematol. 2025 Aug 29

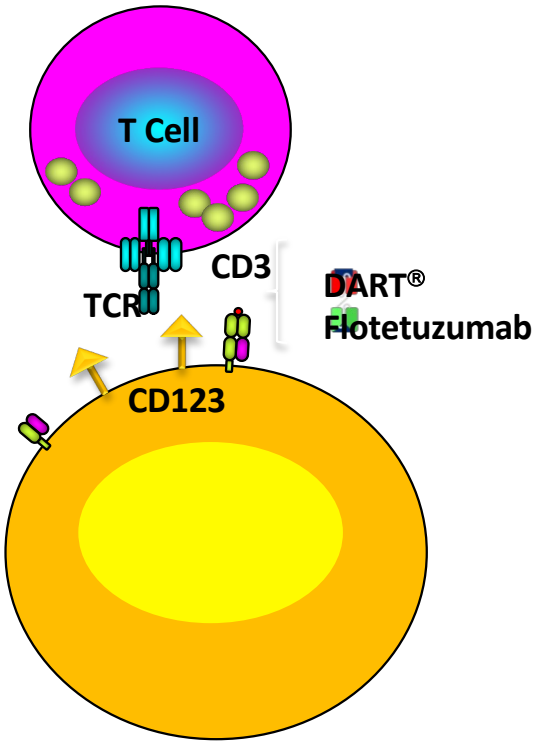
Targeting LSCs and AML blasts by using monoclonal antibodies



Schurch, Front. Oncol., 18 May 2018

Flotetuzumab

CD123 X CD3 Bispecific DART protein

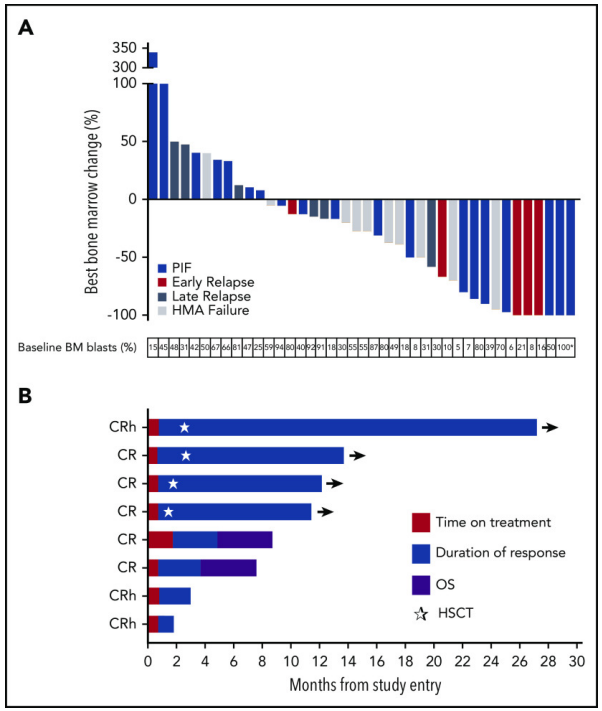


Flotetuzumab is a bispecific molecule that co-engages CD3 T-cell with AML-associated antigen, CD123

Flotetuzumb redirects T cells to lyse CD123-positive AML cells

CD123 is expressed also in normal cells, such as DCs, basophils, monocytes and HSPCs

Flotetuzumab as salvage immunotherapy for refractory AML



Best change in BM blasts, duration of remission, and OS in patients receiving flotetuzumab immunotherapy

Multicenter, open-label, phase 1/2 study of flotetuzumab in 88 adults with relapsed/refractory AML

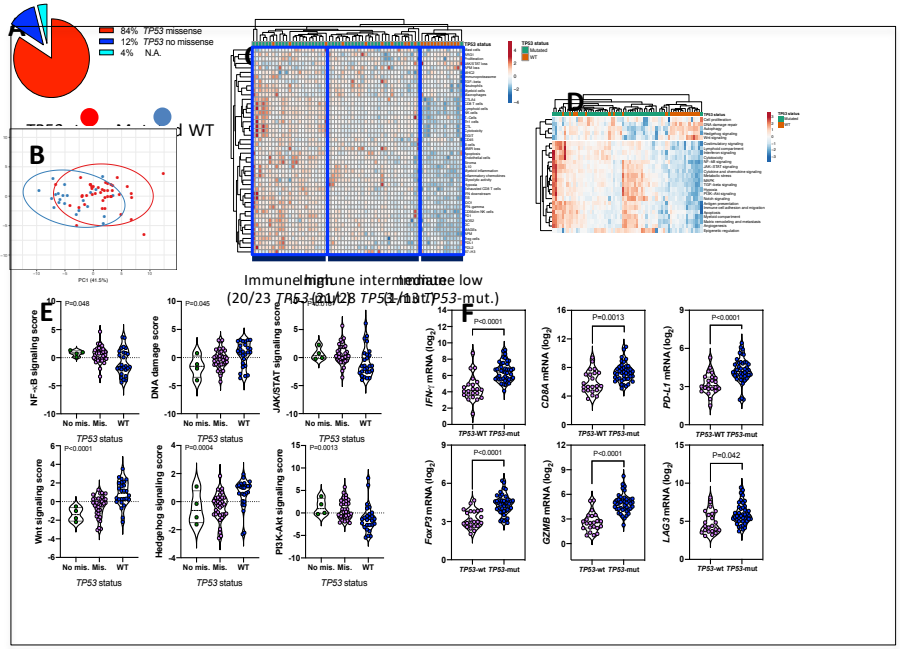
The most frequent adverse events were infusion-related reactions (IRRs)/cytokine release syndrome (CRS), largely grade 1-2

Clinical benefit accrued to PIF/ER patients showing an immune-infiltrated TME

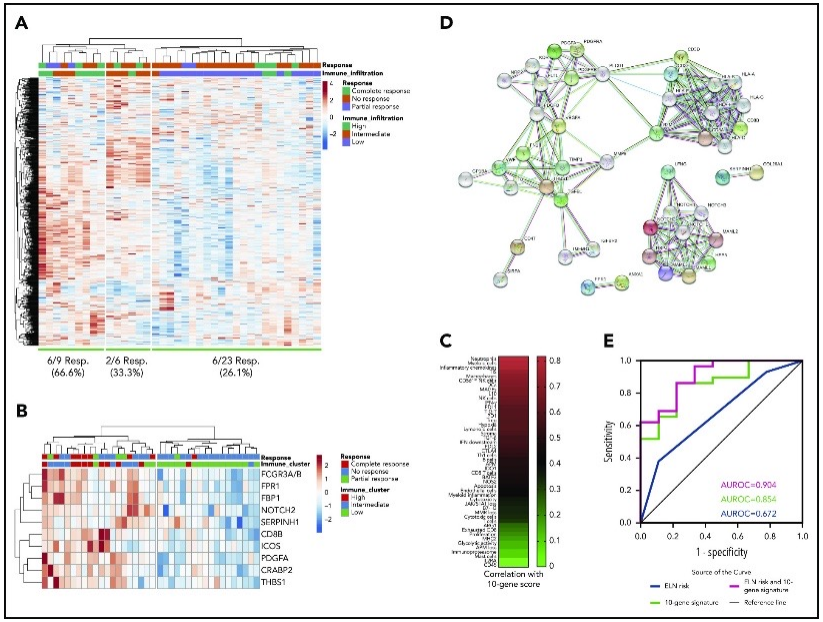
Among 30 PIF/ER patients, the complete remission (CR)/CR with partial hematological recovery (CRh) rate was 26.7%, with an overall response rate (CR/CRh/CR with incomplete hematological recovery) of 30.0%.

Uy GL et al, Blood. 2021 Feb 11; 137(6): 751–762.

TP53 abnormalities correlate with immune infiltration and associate with response to flotetuzumab immunotherapy in AML



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



Uy GL et al, Blood. 2021 Feb 11; 137(6): 751–762.

A 10-gene immune signature predicts response to flotetuzumab with greater accuracy than the ELN risk classifier.

T cell–redirecting therapies in AML

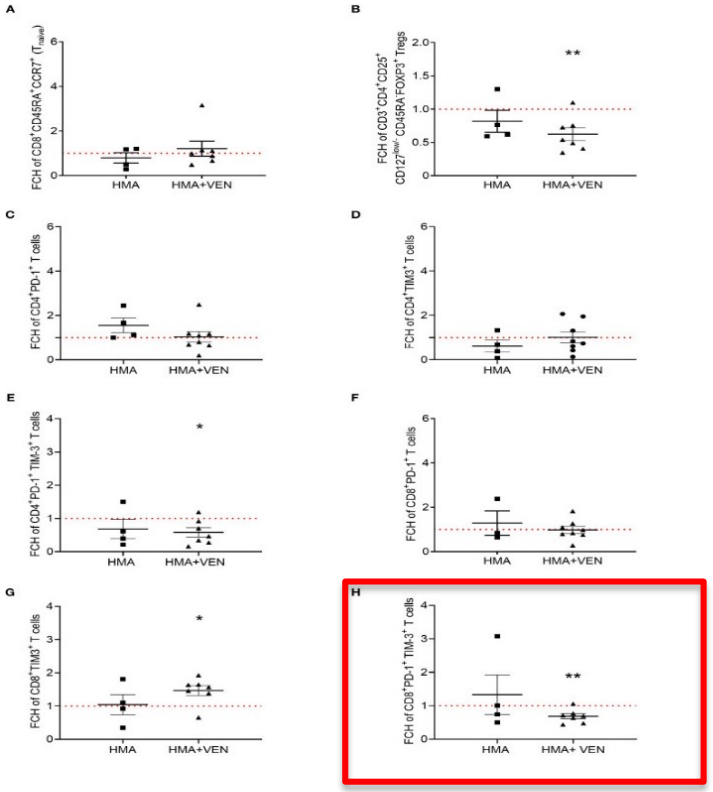
Challenges

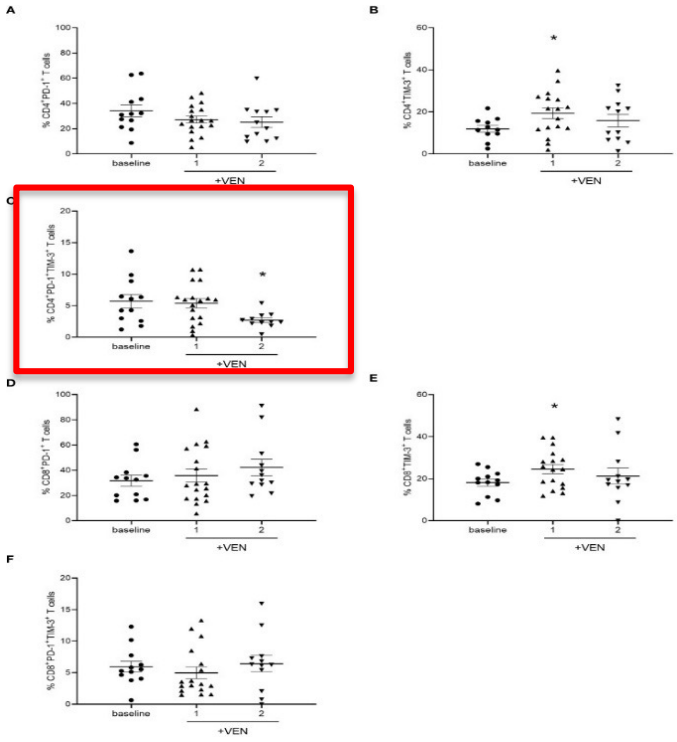
•**High leukemia burden** is associated with severe CRS and limited efficacy
•**Antigen sink** from non-leukemic myeloid cells necessitates higher dosing and increases toxicity risk
•**Step-up dosing** has emerged as essential to reduce CRS risk while preserving efficacy
•**Target antigen promiscuity** remains a major limitation, necessitating dual or tri-antigen targeting and refined patient selection.

How to improve clinical results:

- Biomarker-guided trials
- Rational combinations (e.g., with venetoclax or hypomethylating agents)
- Next-generation constructs with tailored CD3 affinity

Ex vivo characterization of acute myeloid leukemia patients undergoing hypomethylating agents and venetoclax regimen reveals a venetoclax-specific effect on non-suppressive regulatory T cells and *bona fide* PD-1^{+}TIM3^{+} exhausted CD8^{+} T cells





Corradi G et al, Front Immunol. 2024 May 15;15:1386517

CAR-T cells in AML: clinical studies

Concept	Bridge to Transplant	Engineered SCT + CAR T*	Possibly „Stand alone“
Sequence	CAR T/NK => allo SCT (alternatively TCE, moAb)	Target-KO or Epitope edited SCT => CAR T (TCE, moAb)	CAR T/NK
Target Antigen	Myeloid antigens, e.g. CD33, CD123, CLL1, CD117	Engineered stem cell graft, e.g. CD45, CD123, FLT-3, KIT	Restricted expression, e.g. ADGRE2, TCR based, e.g. FLT3-TKD^{mut}, NPM1^{mut}
Hemato-poeitic Toxicity	Yes CART depletion, Stem Cell Salvage	No normal hematopoiesis invisible	Yes / No, depends
CAR	2nd/3rd generation CART, Compound CART	2nd/3rd generation CART, Dual CART	2nd CART, Split CART, „if better“, adapter CART, TCR transgenic T cells
Cell Source	auto or allo T & NK cells, no persistence	donor derived, possibly autologous	patient derived, allo CAR T/NK donor derived post allo SCT
Potential	Improve outcome post SCT; decrease conditioning and thereby increase number of pts eligible for allo SCT	Increase safety by decreasing on-target-off-leukemia toxicity; multiple-targeting possible to overcome antigen escape	Replace allo SCT, applicable to the majority of AML pts
References	Driouk et al, Front Immunol 2020; Zhang et al, Leukemia 2022, Budde et al, Blood 2017; 130, Tambaro et al, Leukemia 2021; Zhang et al, Clin Cancer Res 2021; Liu et al, Blood 2018; 132. Sallman et al, ASH 2022, ASGCT 2023; ASH 2023: Shah et al, #771, Zhang et al, #2106	Kim et al, Cell 2018; Borot F. et al, PNAS 2019; Wellhausen et al, STM 2023; Humbert et al, Leukemia 2019, Chiesa et al, NEJM 2023; Casirati et al, Nature 2023; Marone et al, J Exp Med 2023; Reviews: Kim. Trends in Cancer 2023; Saniei et al, Cell Stem Cell 2023, Volta et al, J Exp Med 2023	Haubner et al, Cancer Cell 2023; Sallman et al, Blood 2019; 134:3826 and Blood 2020;136:(Suppl1); Biernracki etal, JCI 2020, Giannakopoulou et al, Nature Cancer2023 Wermke et al, Blood 2021; Nixdorf et al, Leukemia 2023; Wermke et al, ASH 2023 #3465

* or other immunotherapy tools, e.g. ADCs, TCE

Subklewe M … Curti A, Semin Hematol. 2025 Aug 29

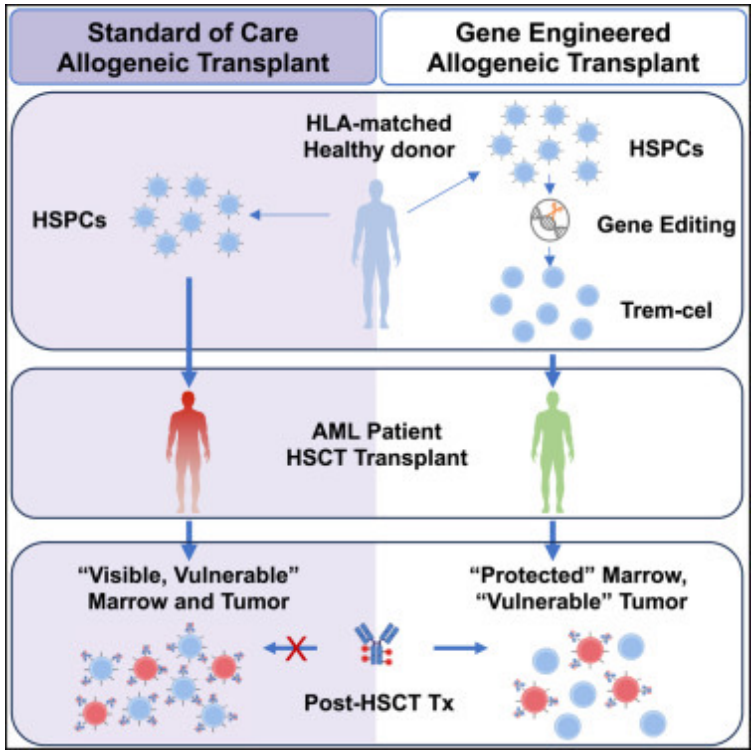
CAR-T cells in AML—What have we learned?

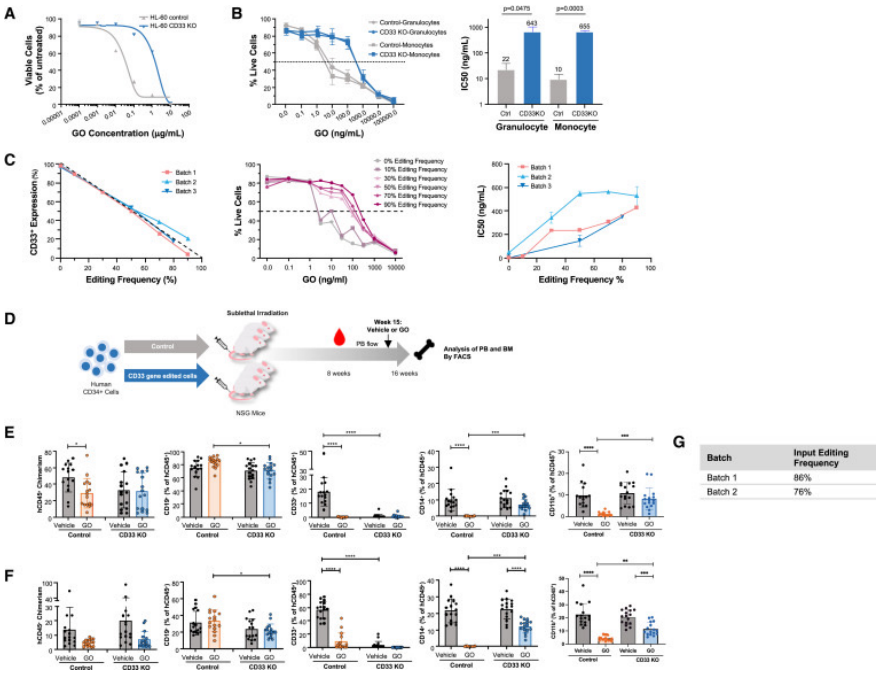
Major challenges:

- Lack of leukemia-specific antigen (on-target, off-leukemia effect)
- Frequent high tumor burden
- Profoundly immunosuppressive bone marrow microenvironment

Most responses observed in early trials have been transient, and long-term remissions remain rare.

Development of a gene edited next-generation hematopoietic cell transplant to enable AML treatment by solving off-tumor toxicity





Lydeard J et al, Mol Ther Methods Clin Dev, 31 (2023)

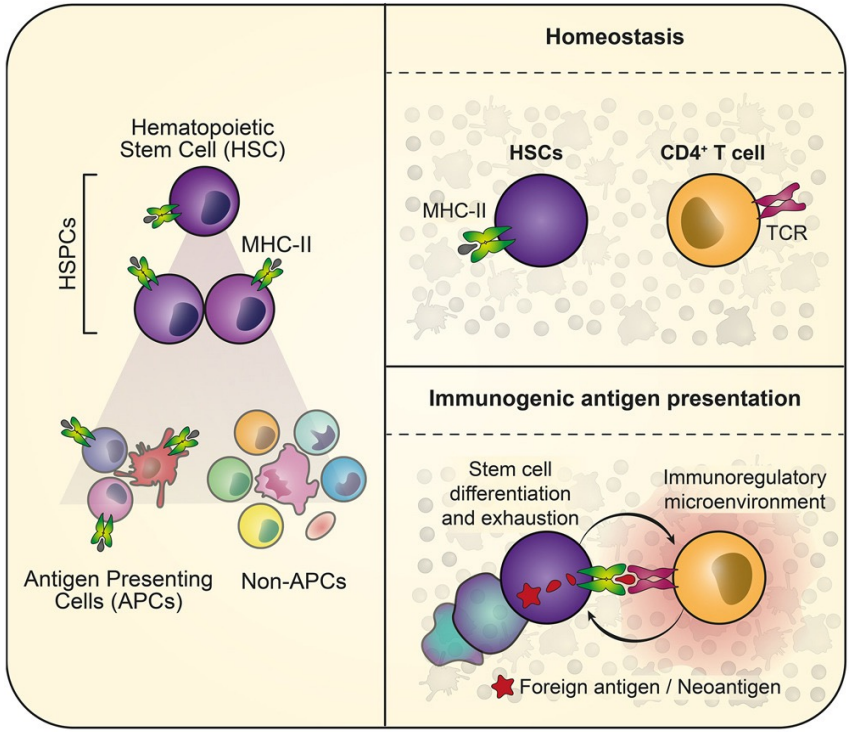
Trial	Patients	Overall response	Response in HR
Aza-Nivo (Phase 2) *Daver et al, 2019*	R/R AML(total 70 pts) HR: 11 ASXL mutated AML	ORR 33% CR/CRi 15% SD (>6 mo) 9%	ASXL1 mut: OR in 7/11 (63%)
HiDAC-Pembro (Phase 2) *Zeidner et al, 2021*	R/R AML(total 37 pts) HR 21 (ELN17)	Primary refractory/early relapse (<6mo) (PIF/ER): PR/ER: 68% ORR 46%; CR/CRi 38%; mOS 13.2 mo	KMT2Ar: CRc in 4/8 (50%) Inv(3)/t(3;3): CRc in 2/4 (50%) ASXL1mut: OR in 3/6 with 1 CR (50%) TP53mut: CRc in 2/5 (40%)
HMA-Sabatolimab (Phase 1b) *Brunner et al, 2021*	ND AML (total 40 patients) 13 pts with molecular HR (TP53/RUNX1/ASXL1)	ORR 40% mDOR 12.6 mo	ORR in 7/13 (53%) mDOR > 12 months
Aza-ven-magro (Phase 3) *Daver et al, 2024*	ND AML (total 189 pts) HR: 44,4%	OS: 11,7 (Magro) vs 10,4 (PBO)	ENHANCE II trial (aza-magro in ND AML): 72 TP53 mutated: ORR 47%; CR/CRi 32%
Aza-durvalumab (Phase II) *Zeidan et al, 2022*	ND AML (total 129 pts) HR: 32 pts (25%; TP53mut 33%	ORR 31% CR 17,2%	RUNX1m had significantly higher ORR
Flotetuzumab (Phase 1/2) *Uy et al, 2021*	R/R AML (total 88 pts) 30 pts PIF/ER; HR 57 pts	In RP2D, CR/CRh rate of 18% (9/50) and an ORR of 24% (12/ 50)	Among PIF/ER pts: ORR 30%; CR/CRh 26,7%, mOS 10,2 mo; ORR 40% in secondary AML
AMG330 (Phase 1) *Ravandi et al, 2024*	R/R AML (total 60 pts) HR: 40 patients (66%)	Among NON responders 37% had at least BM blast reduction >50%	CR/MLFS 13%

HR patients: 297; ORR: 42%

Immunotherapy in AML: potential reasons for reduced efficacy

- AML lacks leukemia-specific antigens. Leukemia mutational burden is relatively low.
- Most targets are shared with normal hematopoietic progenitors, leading to on-target/off-leukemia toxicity.
- AML exerts local and systemic immunosuppression through both tumor-intrinsic and microenvironmental mechanisms, limiting T-cell persistence and function as well as T-cell cytotoxicity.

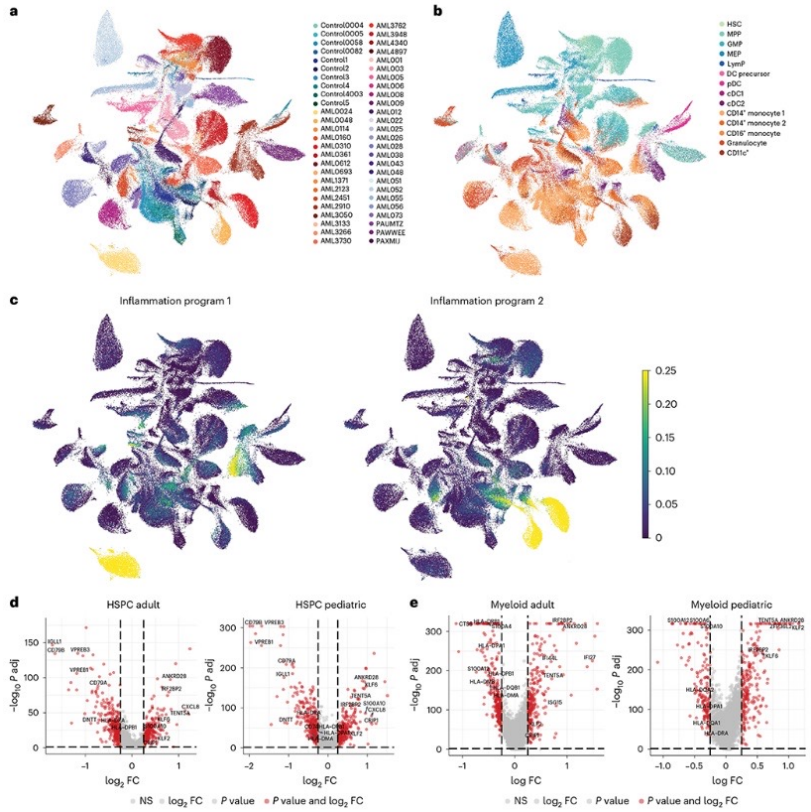
Antigen presentation safeguards the integrity of the hematopoietic stem cell pool

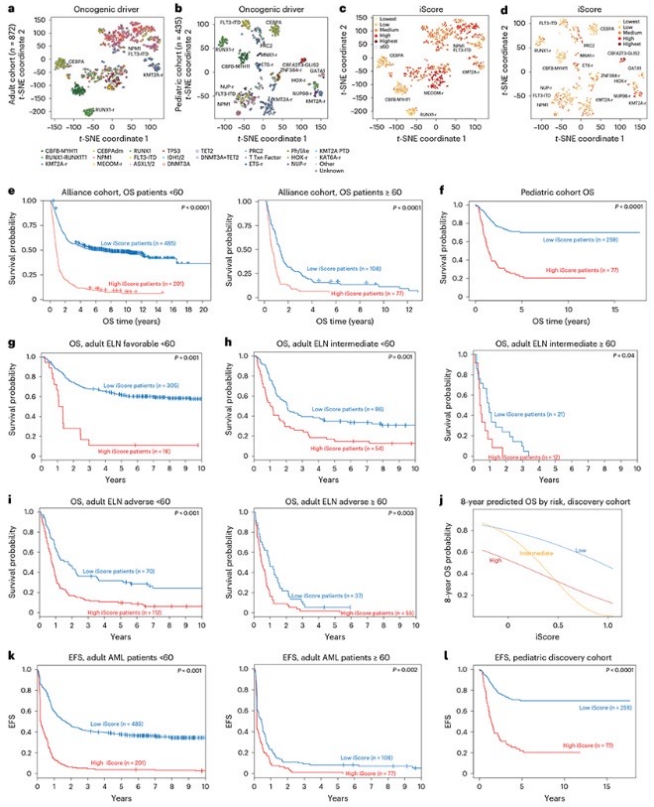


- HSPCs constitutively present antigens via MHC-II
- Presentation of immunogenic antigens results in the activation of $CD4^+$ T cells
- Antigen presentation causes differentiation and depletion of immunogenic HSPCs
- This prohibits the onset of HSC-derived leukemias presenting neoantigens via MHC-II
- $CD4^+$ 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

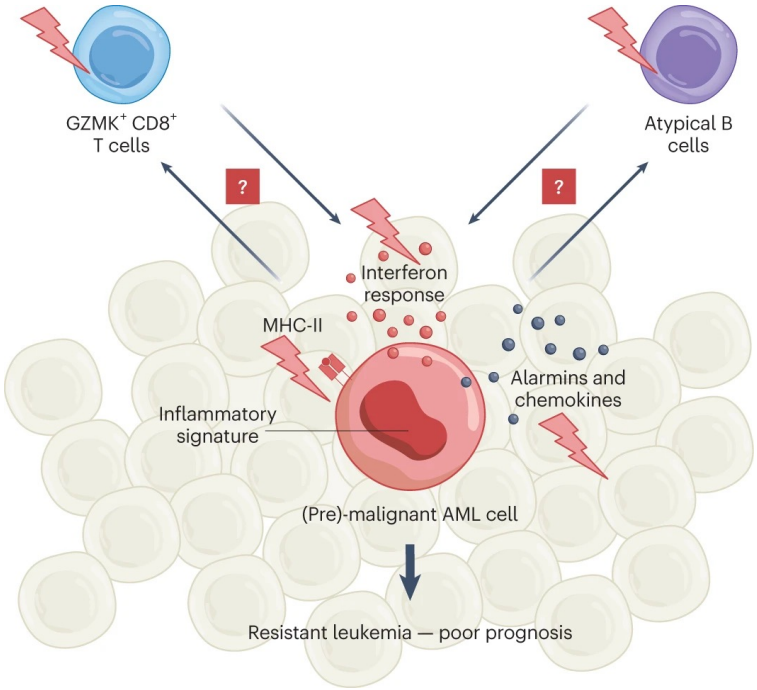
High inflammatory score is associated with adverse ELN risk group and refines prognostic stratification in AML patients





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



Asaf D. Yanir & Shai Izraeli Nature Cancer, 2023, pages 3–4
Commentary on Lasry A et al, Nature Cancer, January 2023, 27-42

Key points

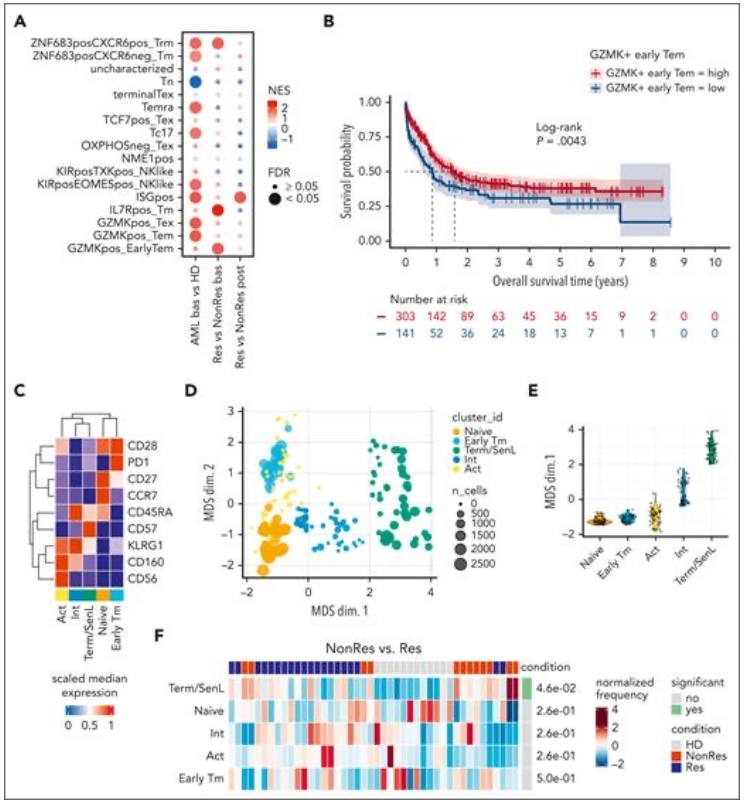
Atypical B cells are associated with inflammation in AML and have suppressive function

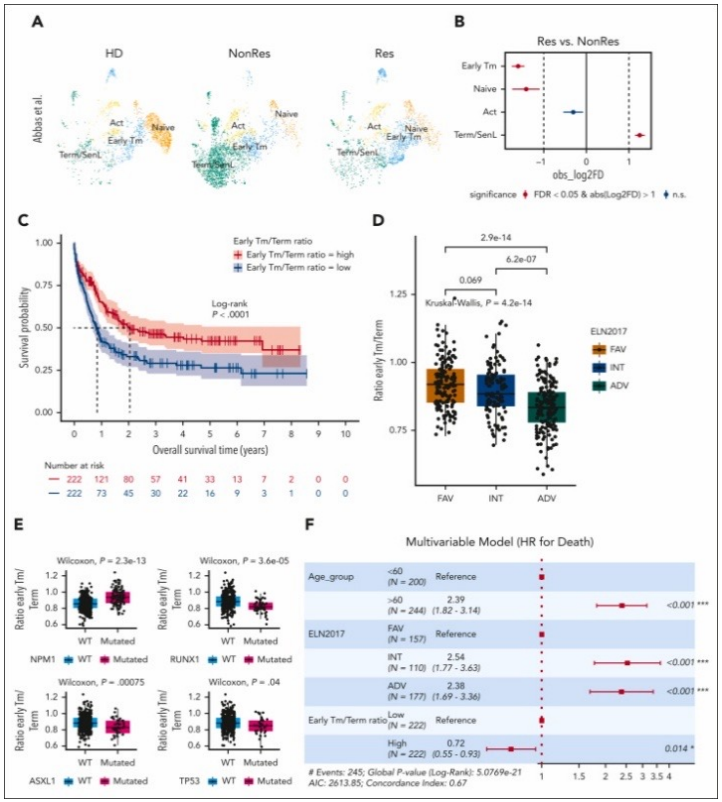
T_{reg} cells and *GZMK*⁺ CD8⁺ T cells are significantly expanded in inflamed patients.

GZMK⁺CD8⁺ T cells are progenitors of terminally exhausted CD8⁺ T (T_{pex}) cells that traffic to sites of inflammation

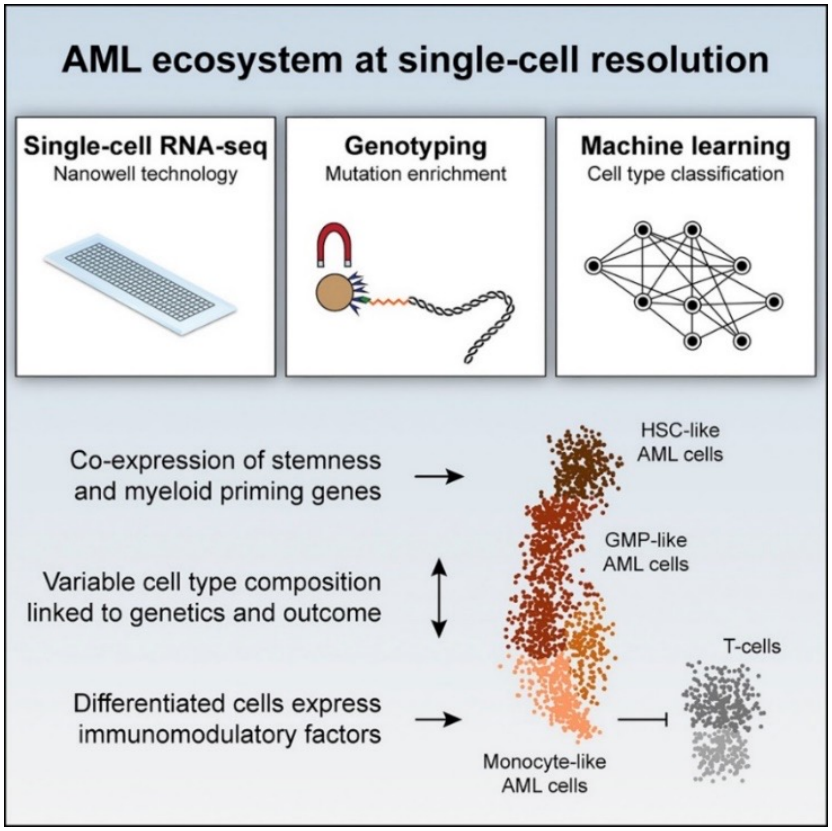
T-cell response is suppressed in high-inflammation patients with AML.

CD8+ T-cell differentiation and dysfunction inform treatment response in acute myeloid leukemia





Mazziotta F et al, Blood. 2024 Sep 12;144(11):1168-1182



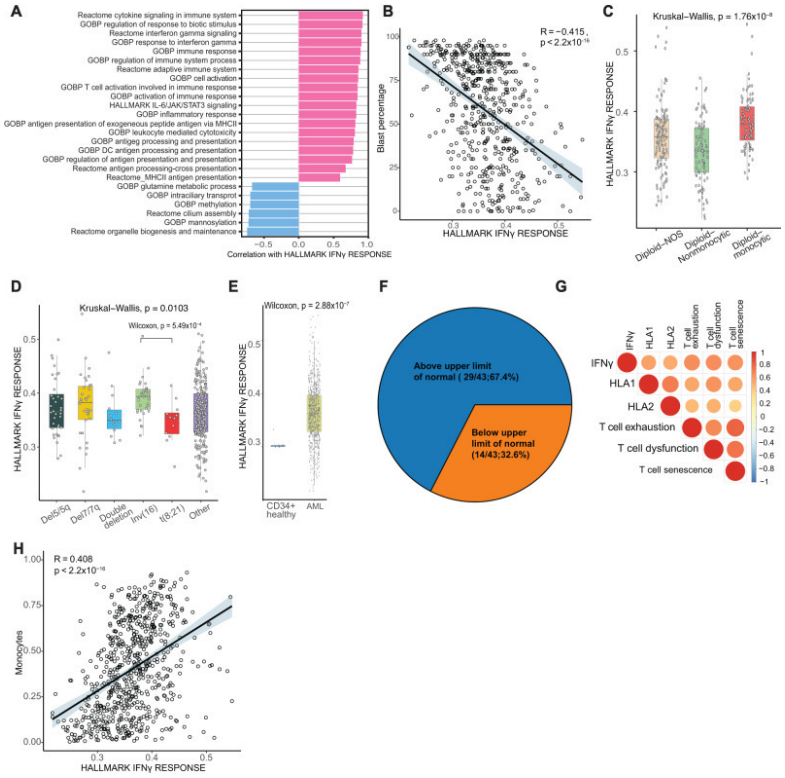
Single-Cell RNA-seq reveals AML hierarchies relevant to disease progression and immunity

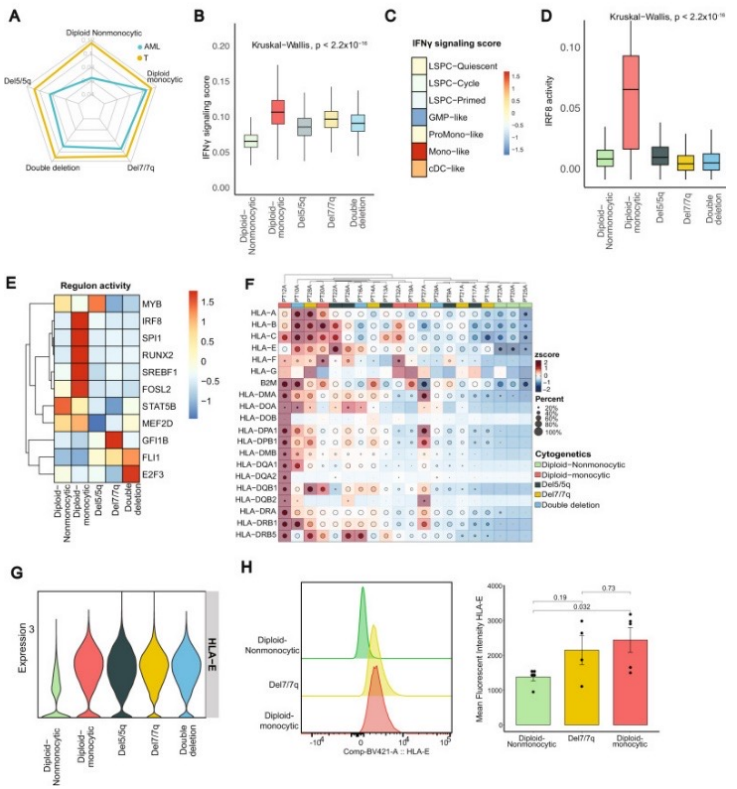
Recent studies using single cell sequencing have revealed the clonal diversity and phenotypic heterogeneity in AML with greater precision.

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

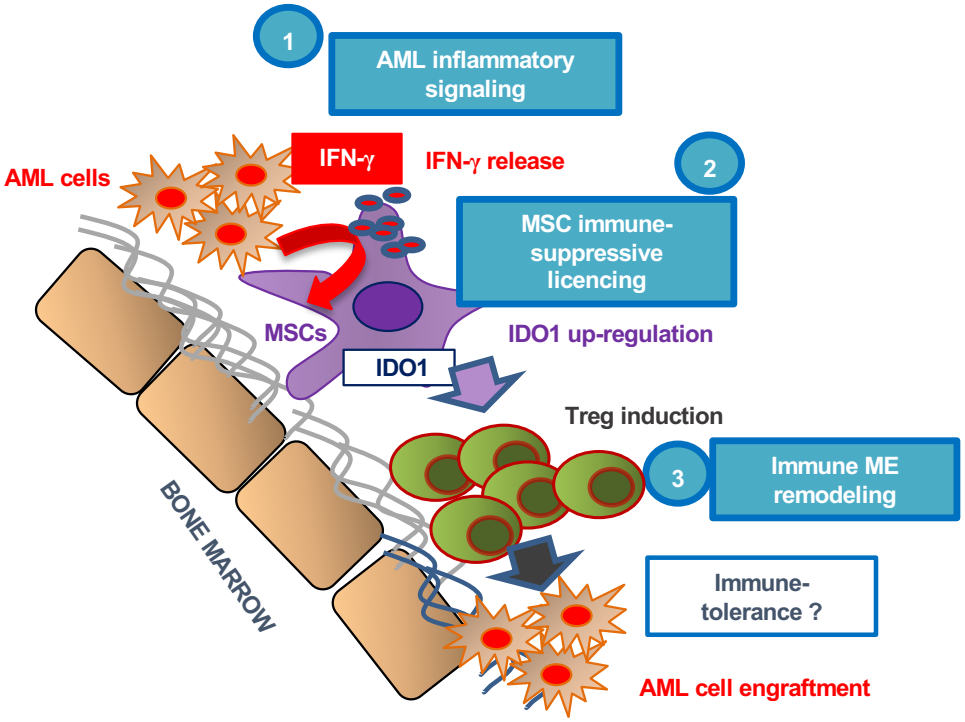
Comprehensive characterization of IFNγ signaling in acute myeloid leukemia reveals prognostic and therapeutic strategies

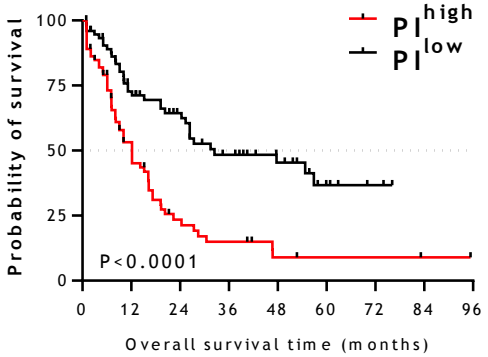


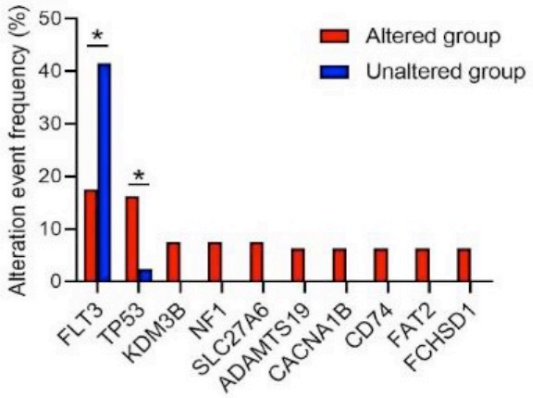


Wang B et al, Nat Commun. 2024 Feb 28;15:1821

IFN-γ-dependent signals produced by AML cells modify MSC functions and favor an immune-modulating milieu

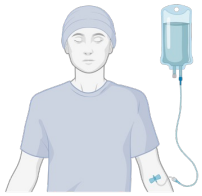




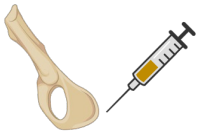


Corradi et al, Clin Cancer Res. 2022 Jul 15;28(14):3141-3155

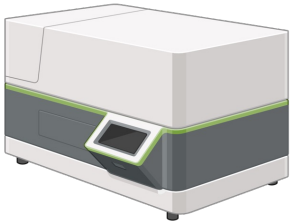
Prognostic Immune Subgroups of Newly-Diagnosed AML



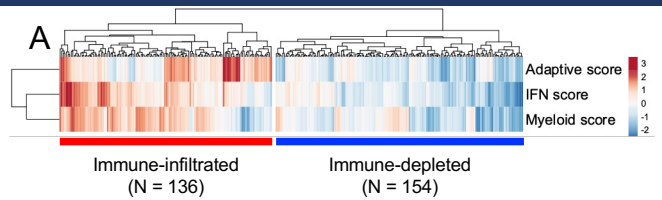
Discovery Cohort (N = 290 ND-AML)
Princess Margaret Cancer Centre, Toronto (Dr. Mark M. Minden)
3+7 Backbone

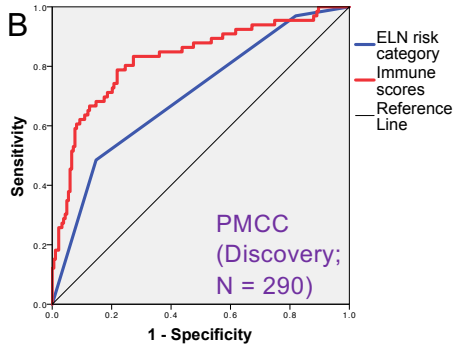


Diagnostic BM Aspirate / Biopsy

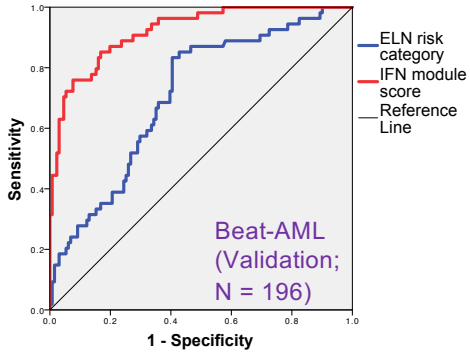


nCounter® Assay
(N = 770 Immune Genes)
+
GeoMx® Digital Spatial Profiling



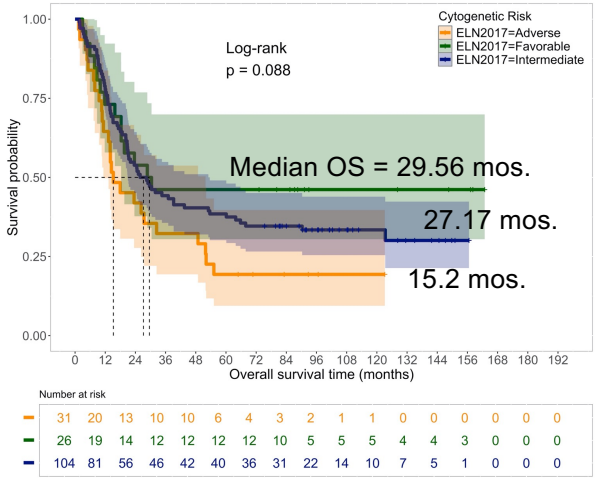


Variable	AUROC	SE	95% CI
IFN scores	0.815	0.031	0.755-0.876
ELN risk	0.702	0.038	0.628-0.776



Variable	AUROC	SE	95% CI
IFN scores	0.921	0.04	0.88-0.961
ELN risk	0.709	0.021	0.629-0.788

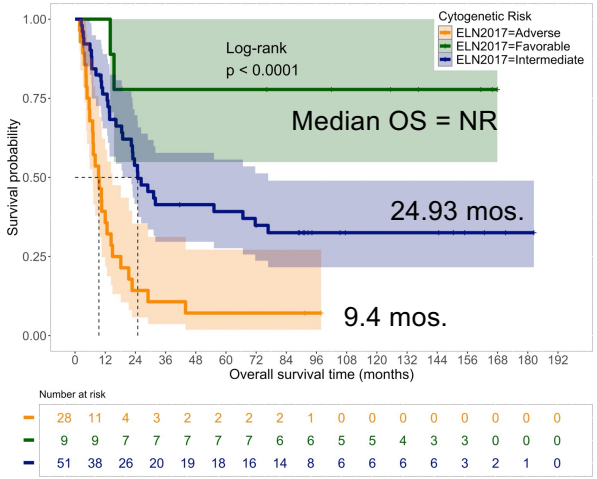
C Immune Subtypes Stratified by ELN2017 Category



Immune-depleted

OS Rates by ELN2017 Risk

	1y	2y	5y
Favorable	73%	54%	46%
Intermediate	78%	54%	39%
Adverse	64%	42%	19%



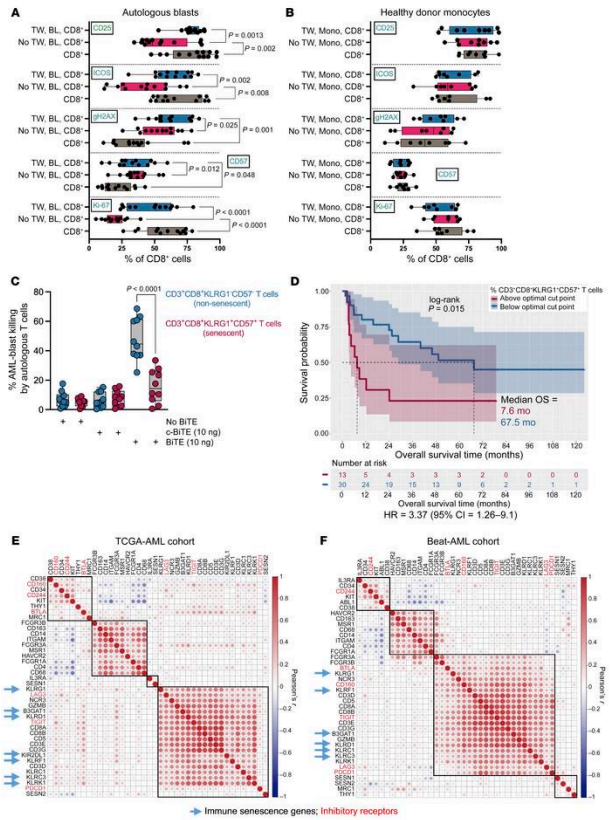
Immune-infiltrated

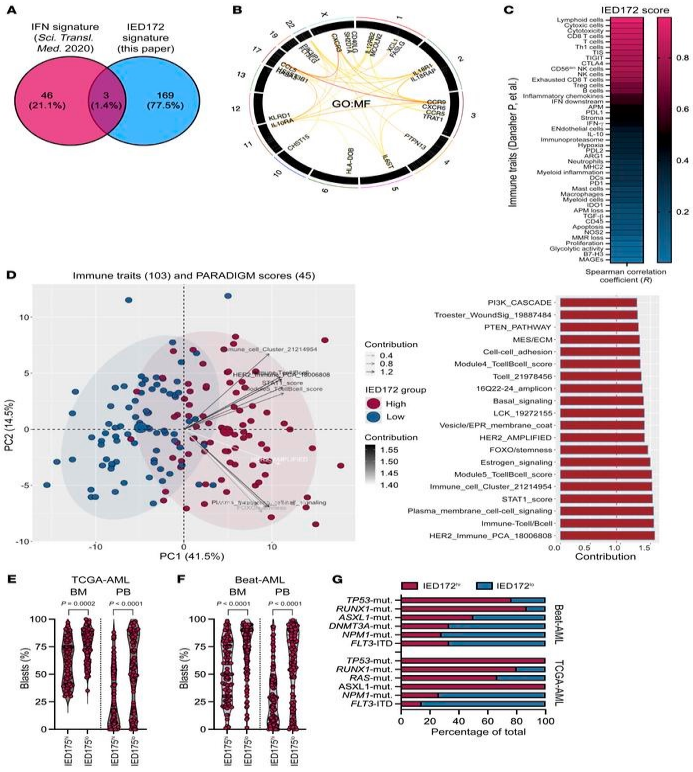
OS Rates by ELN2017 Risk

	1y	2y	5y
Favorable	100%	78%	78%
Intermediate	76%	54%	39%
Adverse	36%	14%	7%

Vadakekolathu J, *et al*. Science Translational Medicine, 2020.

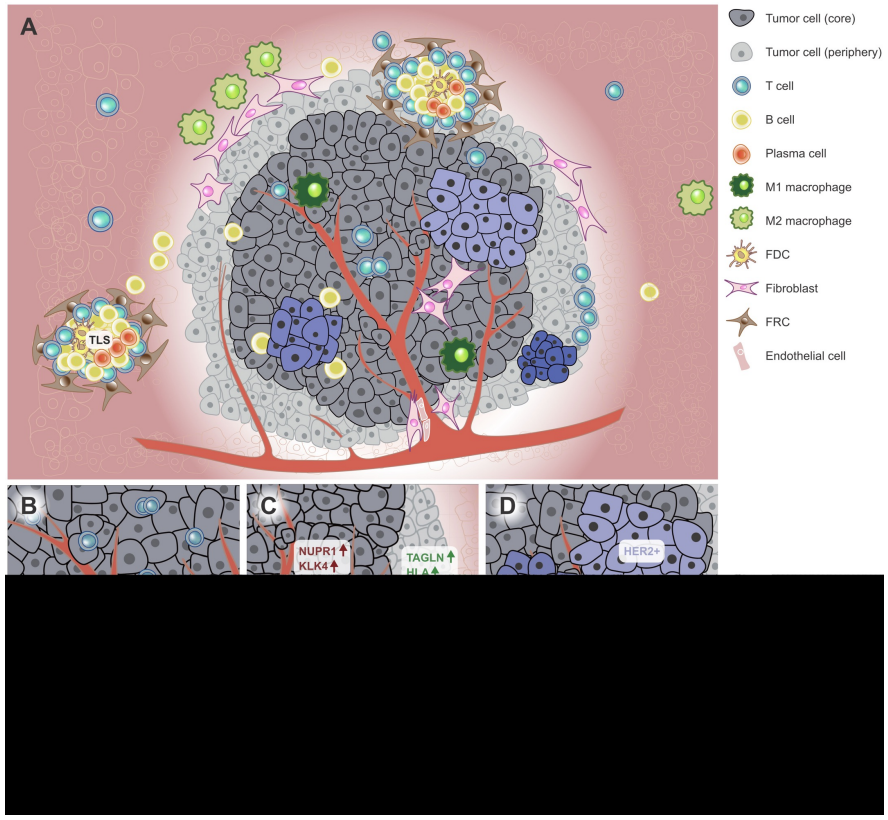
Signatures of immune effector dysfunction and T cell senescence correlate with immune infiltration/inflammation and response to immune checkpoint inhbition





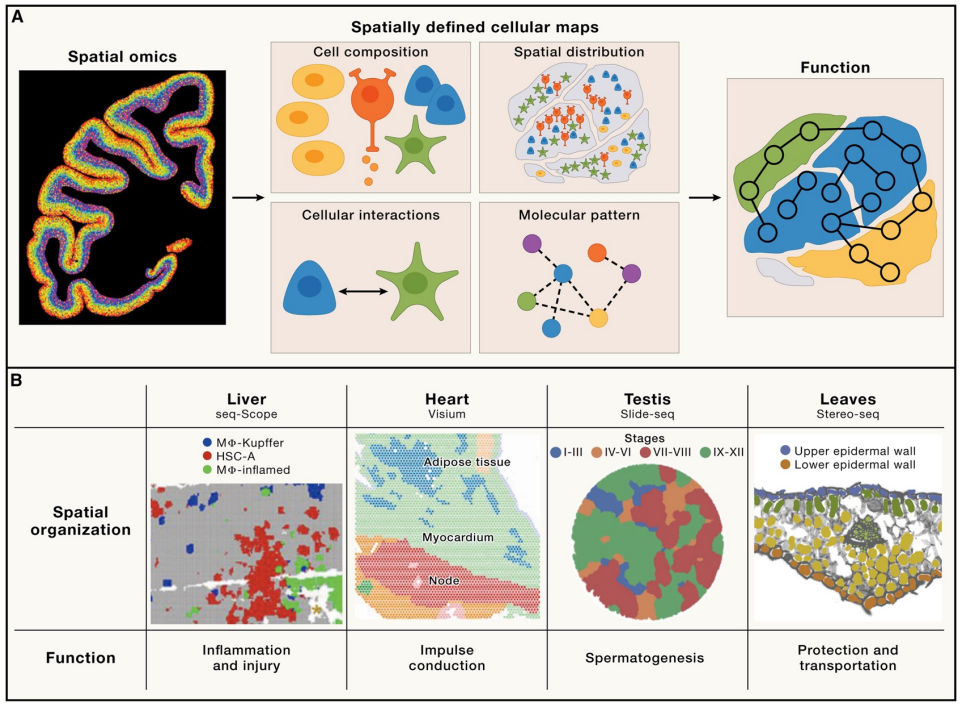
Rutella S et al, J Clin Invest, sept 13, 2022

Spatial multi-omics as a novel platform for immunotherapy



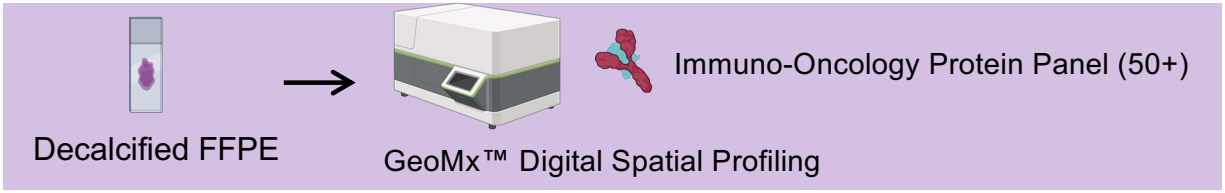
Yu Q et al, Front. Oncol., 13 October 2022

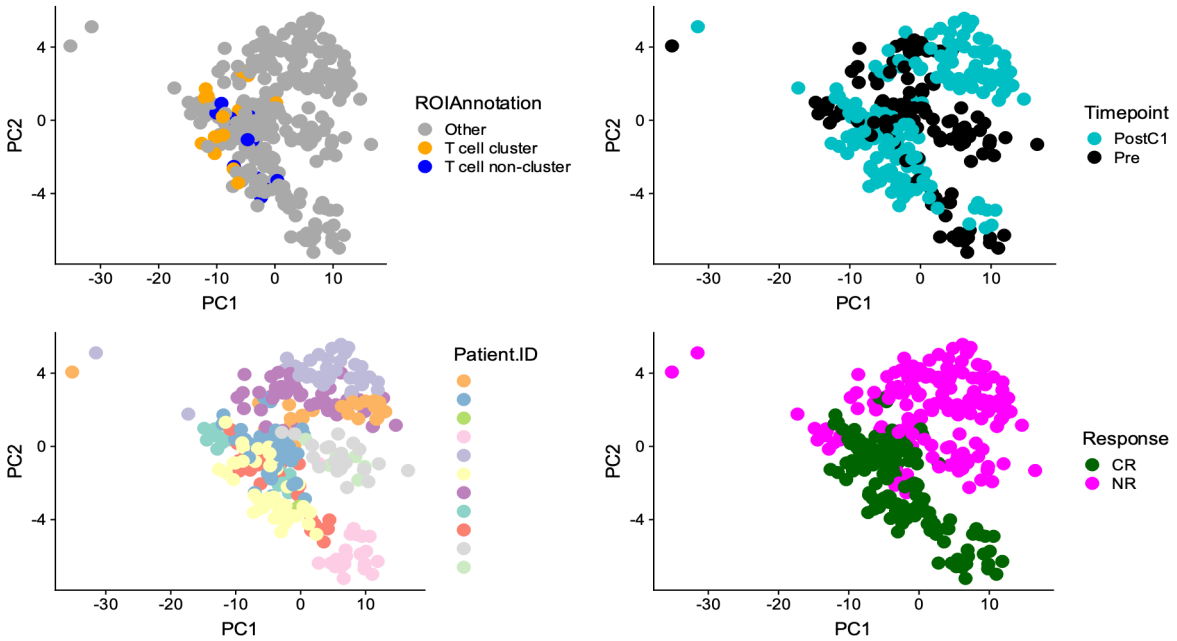
Spatiotemporal omics for biology and medicine



Liu L et al, Cell, Volume 187, Issue 17p4488-4519 August 22, 2024

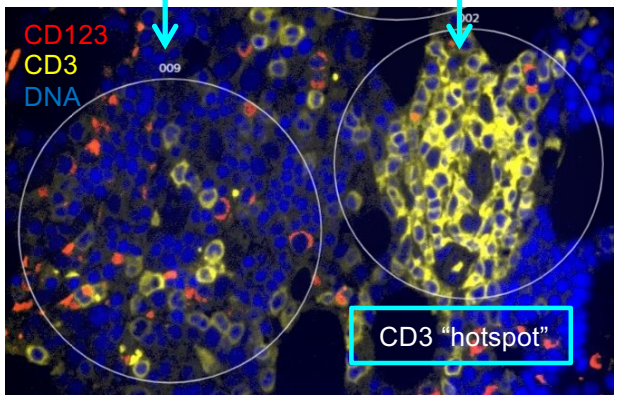
Spatial Proteomics Reveals T-Cell Activation Within Immune ‘ Hotspots

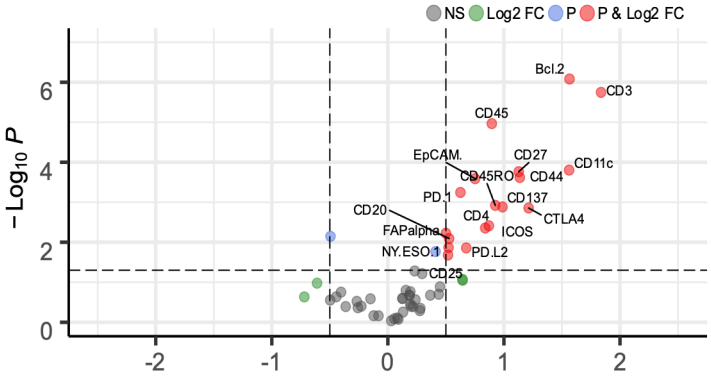




Region of interest (ROI) without T-cell clustering around CD123⁺ AML cells

ROI with T-cell clustering around CD123⁺ AML cells





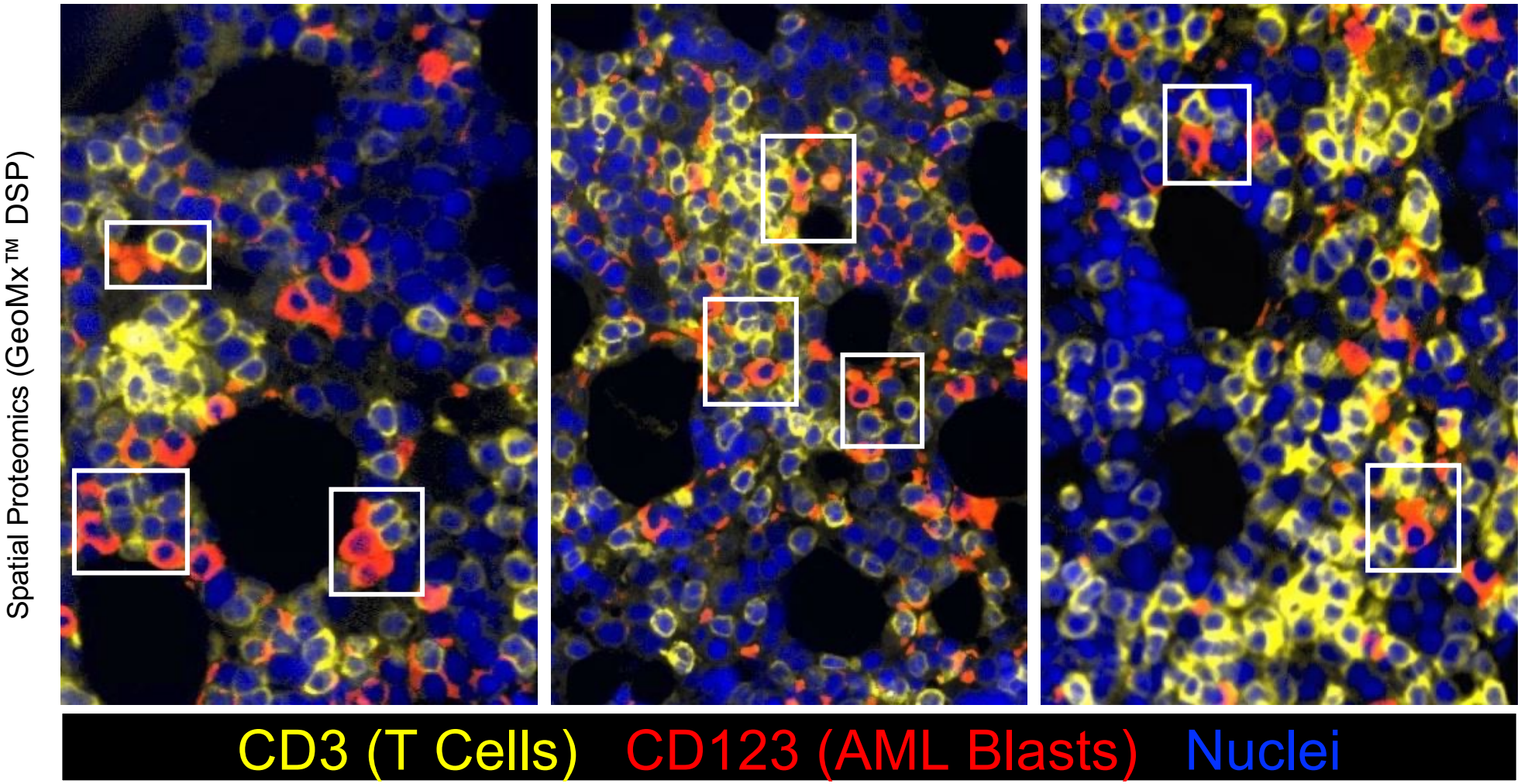
Low in ROIs with T-cell ‘hotspots’

High in ROIs with T-cell ‘hotspots’

Created with BioRender.com

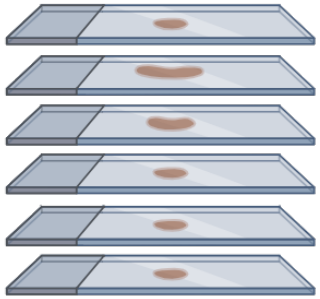
Vadakekolathu J, *et al*. Science Translational Medicine, 2020 and Unpublished.

Putative Immunological Synapses in Flotetuzumab Responders



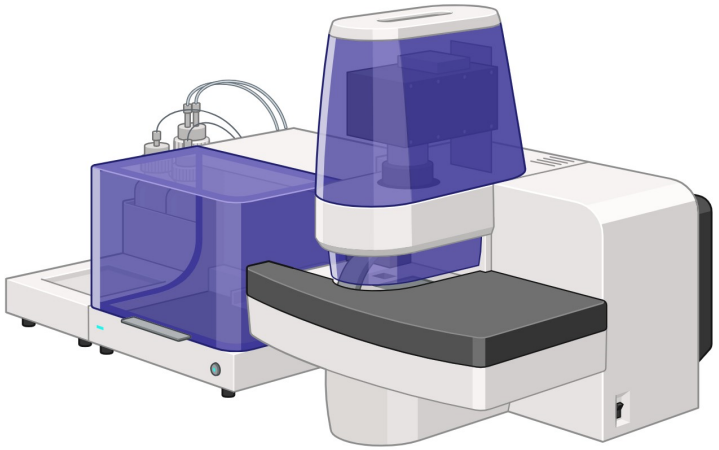
Towards spatially-resolved tumor microenvironment heterogeneity from tissue biopsies in AML

AML patients at the diagnosis
N=6 (FFPE)



BoneSTATION (Milestone)

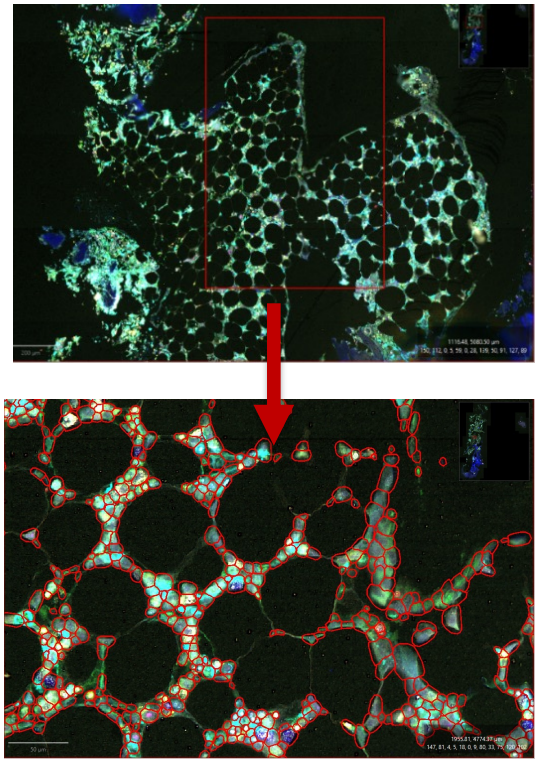
Phenocylcer-FUSION (Akoya Biosciences)



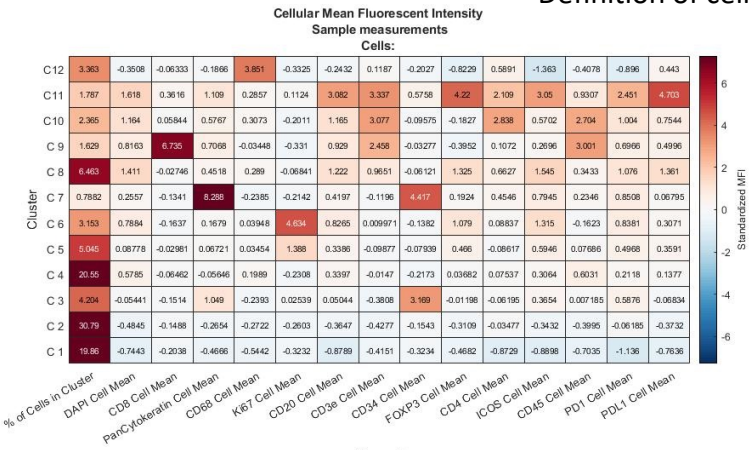
- Bone marrow processing within 48 hours
- Decalcification process reduction
- MoL-DECALCIFIER: innovative pure EDTA solution without requiring an additional acid/base buffer

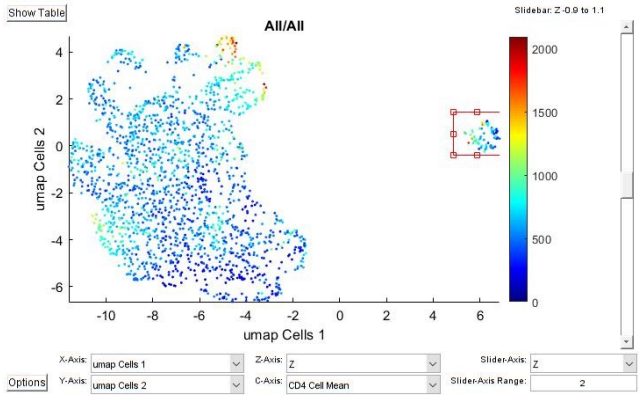
Spatial proteomic analysis in AML patients at diagnosis

Segmentation

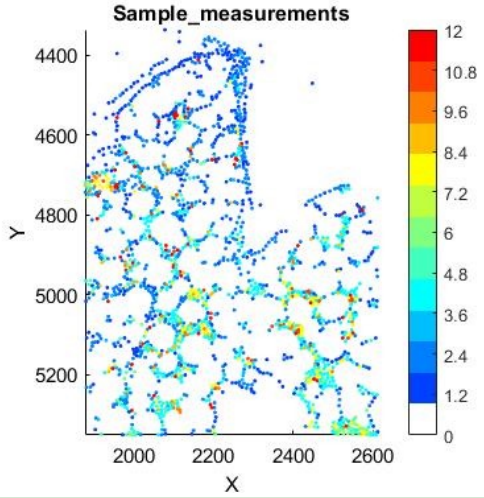


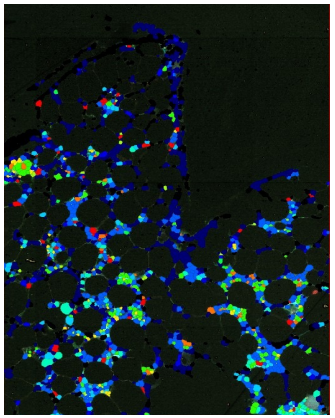
Definition of cell subtypes and clusters





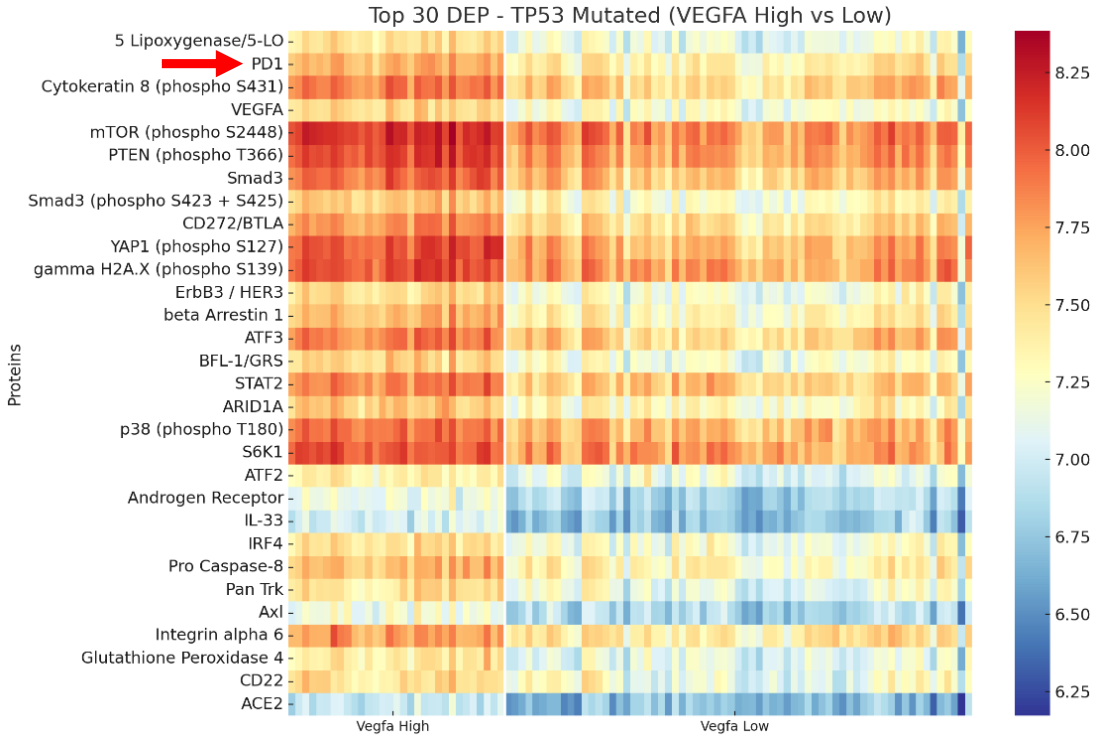
Channel

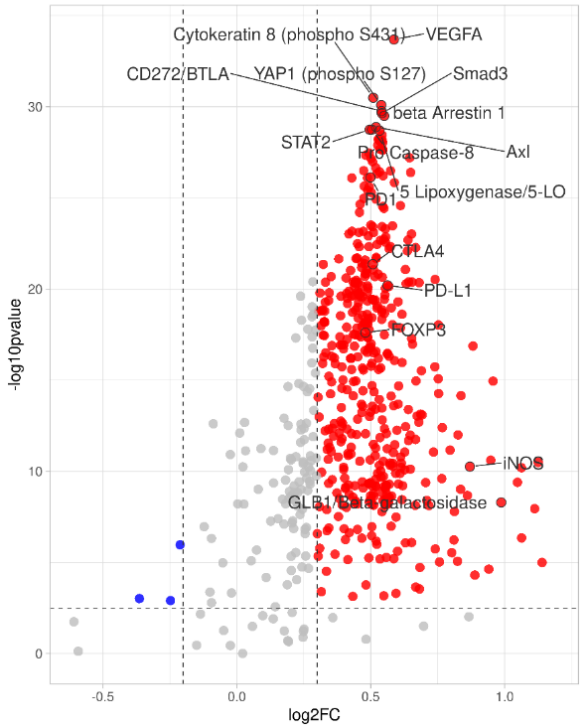




Spatial proteomic analysis with unsupervised clustering using QuPath and CytoMAP

Loco-regional VEGFA concentration is associated with an immune-suppressive microenvironment in TP53 mutant AML patients





VEGFA low VEGFA high

Unpublished data

Conclusions and Perspectives

- Despite extraordinary progress in the treatment of lymphoid malignancies through immunotherapy, AML has proven far more resistant to this revolution.
- While AML may not yet have had its immunotherapy revolution, the knowledge how to improve safety and efficacy is growing. Advances in spatial biology, single-cell profiling, and proteogenomics, will help refine patient selection and treatment sequencing.
- The way forward is not a one-size-fits-all solution but a personalized immunotherapeutic strategy that adapts to each patient's leukemic and immunological landscape..
- The future of AML immunotherapy will depend not only on better target antigen selection and engineering of more potent effector platforms, but also on integrating immune-based treatments with established backbones such as azacitidine/venetoclax and hematopoietic stem cell transplantation.

HIGHLIGHTS IN EMATOLOGIA

TREVISO, 7-8 NOVEMBRE 2025

Seragnoli Institute

Acute Leukemia and MDS Group

Laboratory	***Clinical Team***	***Study coordinators***
Valentina Salvestrini	Cristina Papayannidis	Francesco Ingletto
Marilena Ciciarello	Stefania Paolini	Manuel Cella
Darina Ocadlikova	Sarah Parisi	Antonella Pagano
Dorian Forte	Chiara Sartor	Gaida Iannichini
Karyna Volkava	Jacopo Nanni	
Federico Pasini	Federico Zingarelli	
Lucia Rossi	Andrea Davide Romagnoli	
Ismael Polanco Sanchez	Raffaele Ciruolo	
	Caterina Azzimondi	

Collaborators:

Roberto Maria Pellegrino
(University of Perugia)

Sabina Sangaletti
Mario Colombo
(INT Milan)

Sharham Kordasti
King's College, London

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

Maura Rossi, Claudio Agostinelli,
Elena Sabattini
(IRCCS AOU Bologna)

Francesco Fabbri, Giorgia Simonetti
(IRST Meldola)

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

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