9° WORKSHOP IN EMATOLOGIA TRASLAZIONALE

DELLA SOCIETÀ ITALIANA DI EMATOLOGIA SPERIMENTALE

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Mechanisms involved in the resistance to anti-CD38 monoclonal antibodies in multiple myeloma

Paola Storti

Dipartimento di Medicina e Chirurgia

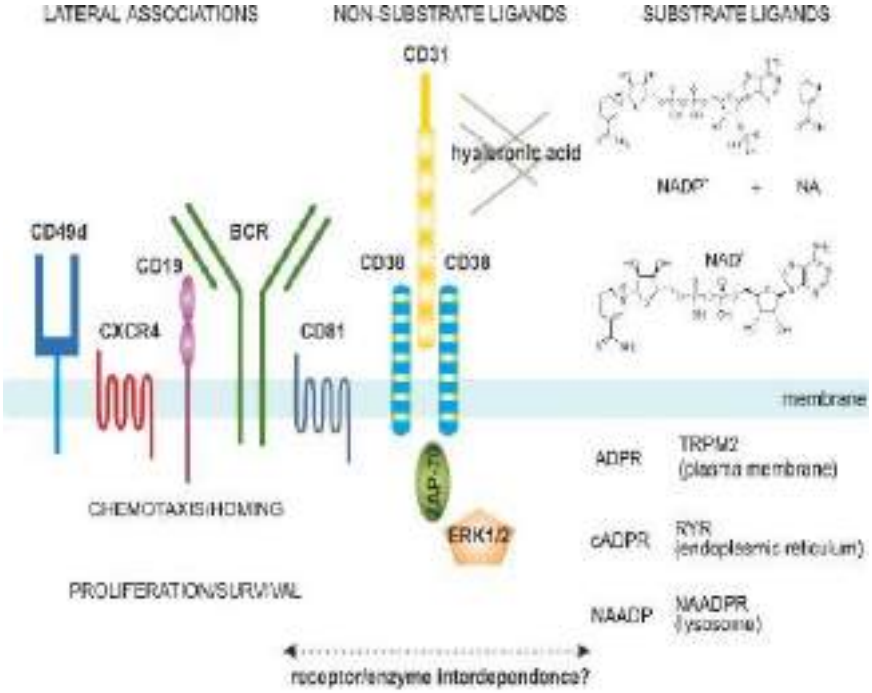
Università di Parma

UNIVERSITÀ DI PARMA

Disclosures di Paola Storti

Company name	Research support	Employee	Consultant	Stockholder	Speakers bureau	Advisory board	Other
Sanofi							x

CD38 is a Cell-surface Receptor and Ectoenzyme



As a receptor

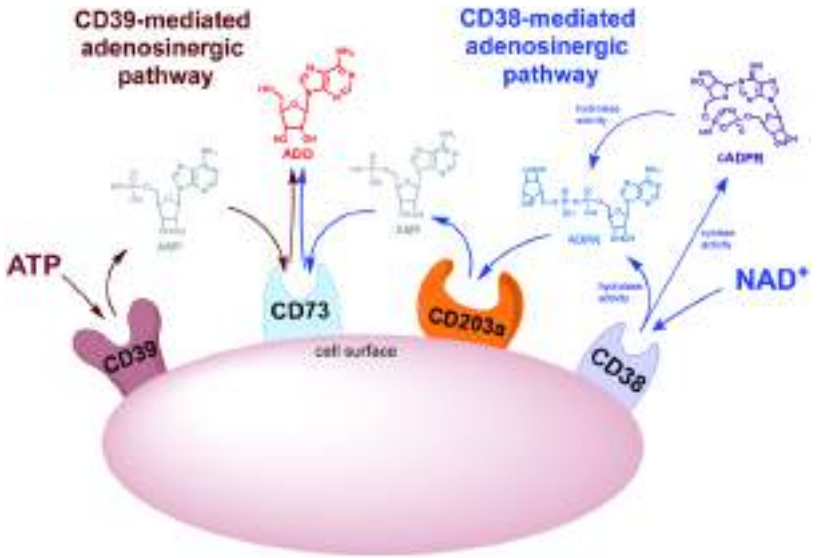
- Regulates signaling, homing, adhesion and migration in close contact with BCR complex and CXCR4.
- Engagement with CD31 or hyaluronic acid activate ZAP-70, ERK1/2 and NFKB pathways and regulates activation and proliferation of the cell.

As an ectoenzyme

- CD38 interacts with NAD+ and NADP+, which are converted to cADPR, ADPR, and NAADP, all intracellular $Ca2^{+}$ mobilizing agents.

Malavasi F et al. *Blood*. 2011 Sep 29;118(13):3470-8

Role of CD38 in Adenosine (ADO) generation



- There are two adenosinergic pathways associated with exogenous ADO generation.
- The better-known pathway involves the nucleoside triphosphate diphosphohydrolase known as CD39.
- A lesser known adenosinergic pathway is mediated by CD38, which hydrolyzes NAD+ to ADPR. ADPR is in turn converted to ADO by the CD203a and CD73.
- Extracellular ADO, which is prominent in the TME, stimulates the ADO receptor, A2AR, on the surface of immune effector cells.
- Collectively these transformations cause a shift from an ATP-driven proinflammatory environment to an anti-inflammatory milieu induced by ADO-mediated down regulation of immune function.

Benton TZ, et al. RSC Adv. 2021

CD38 expression profiles

Lymphoid tissue	Cell population
Blood	T cells (precursors, activated) B cells (precursors, activated) Myeloid cells (monocytes, macrophages, dendritic cells) NK cells Erythrocytes Platelets
Cord blood	T and B lymphocytes, monocytes
Bone marrow	Precursors Plasma cells
Thymus	Cortical thymocytes
Lymph nodes	Germinal center B cells

- **Highly and uniformly expressed on multiple myeloma (MM) cells**[1,2,3]
- **Relatively low expression on normal lymphoid and myeloid cells** and in some tissues of non-hematopoietic origin[4]
- **CD38 is not expressed on hematopoietic pluripotent cells**, which are crucial for the recovery of the long-term bone marrow

Malavasi F et al. *Physiol Rev*, 2008; Lin P et al. *Am J Clin Pathol*, 2004; Santonocito AM et al. *Leuk Res*, 2004; Deaglio S et al. *Leuk Res*, 2001.

Rationale for targeting CD38

Functions:

1) Receptor-mediated adhesion and signaling functions
2) Enzymatic activities

   Contributes to intracellular calcium mobilization

   Involved in production of adenosine: important for induction of local immunological tolerance → implicated in local survival strategy of the neoplastic plasma cell in the bone marrow milieu

Expression levels:

1) Low level of expression of CD38 on lymphoid and myeloid cells under normal conditions
2) High level of CD38 expression on malignant cells in MM

De Weers M et al. *J Immunol,* 2011; Chillemi A et al. *Mol Med,* 2013; Quarona V et al. *Ann N Y Acad Sci,* 2015.

Two anti-CD38 mAbs clinical approved

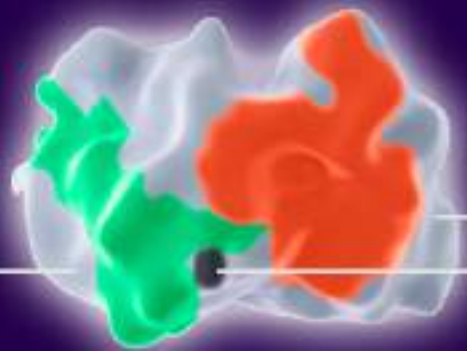
Isatuximab is an IgG1-κ chimeric, humanized anti-CD38 monoclonal antibody that binds selectively to a specific epitope on the cell surface antigen CD38, opposite to the catalytic site[2]

Daratumumab is an IgG1-κ humanized anti-CD38 monoclonal antibody, which targets a specific discontinuous region on CD38 that includes residues located opposite to the active site[2]

CD38

The epitopes of human CD38 (huCD38) interacting with isatuximab and daratumumab are distinct[1]

Isatuximab and daratumumab induce different structural changes within the CD38 molecule upon binding[1,2]



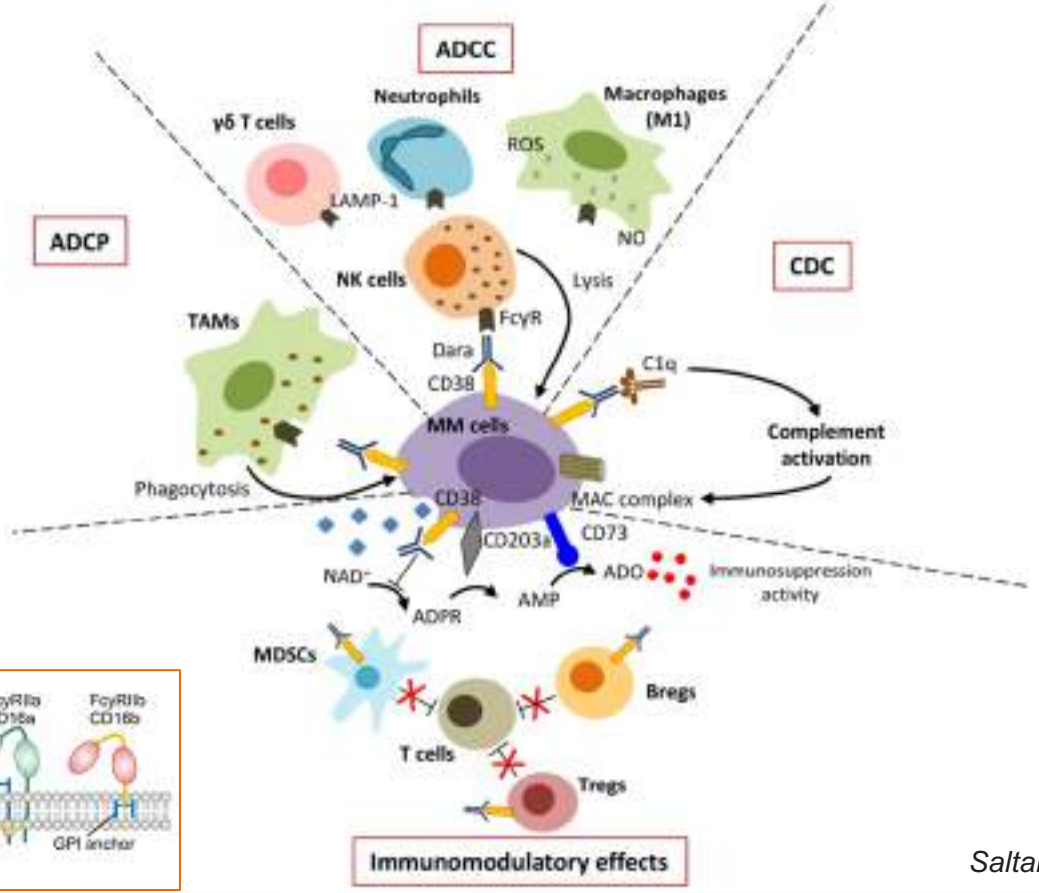
Isatuximab epitope

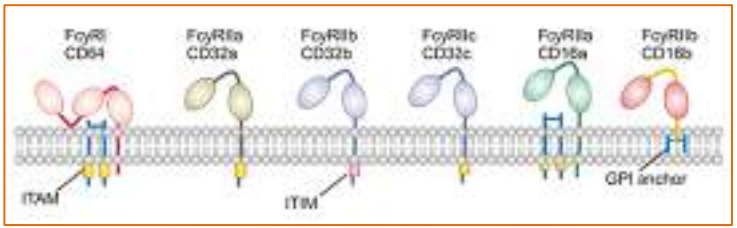
One shared amino acid (Glu233)

Daratumumab epitope

Martin TG et al. Cells 2019 Nov 26;8(12):1522

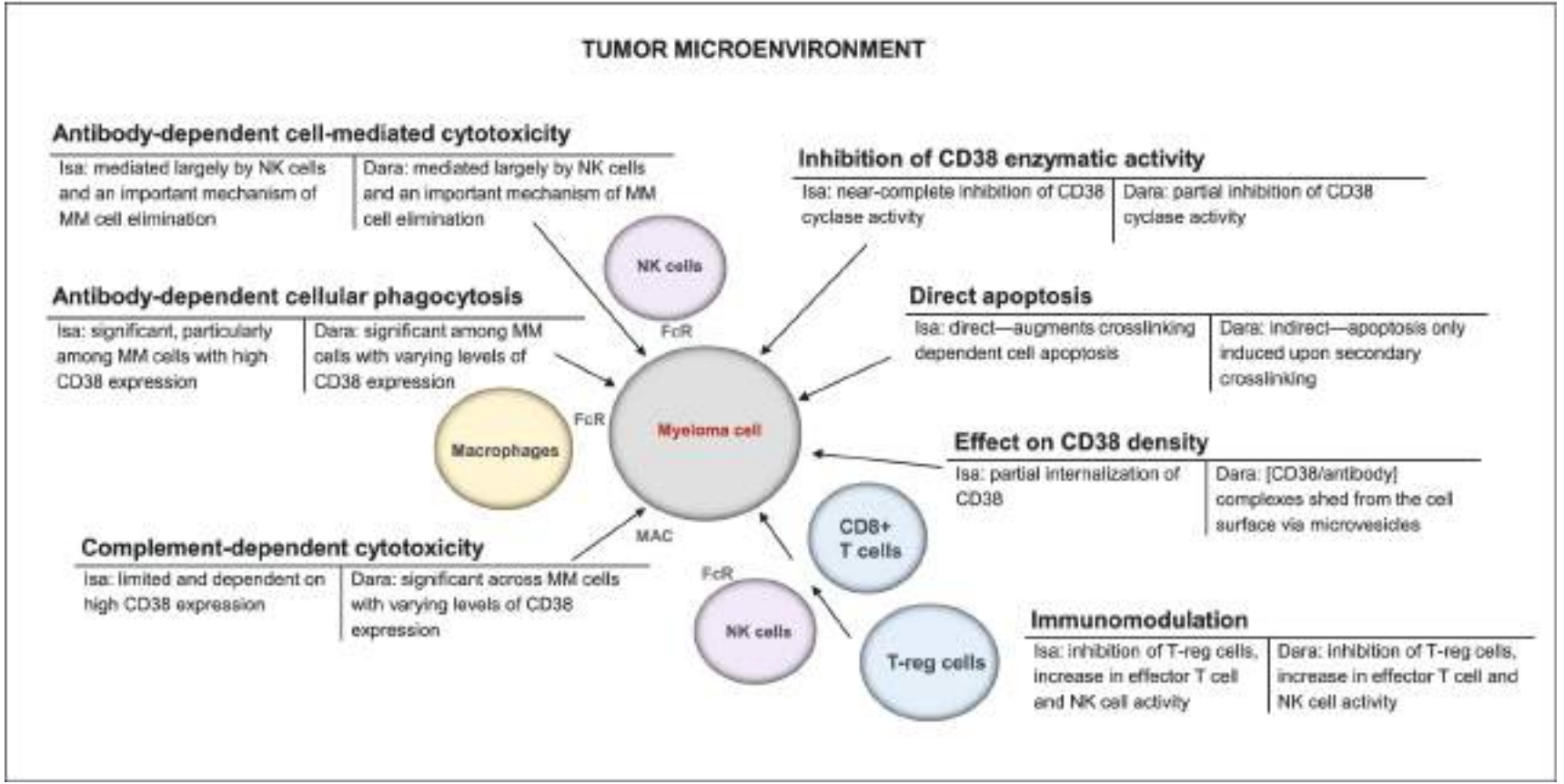
Mechanisms of actions of anti-CD38 mAbs





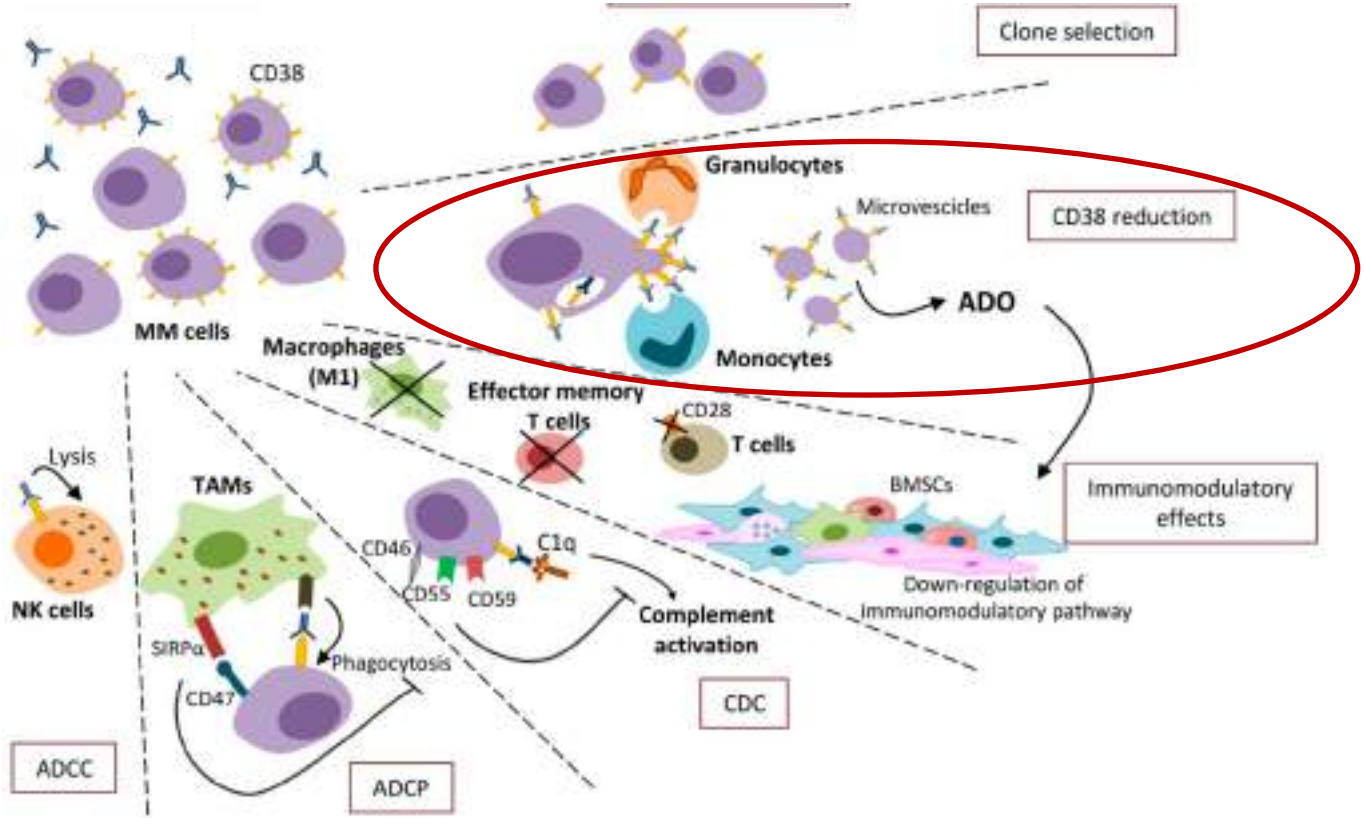
Saltarella I., Cells **2020**

Different mechanisms of action of the two anti-CD38 mABs



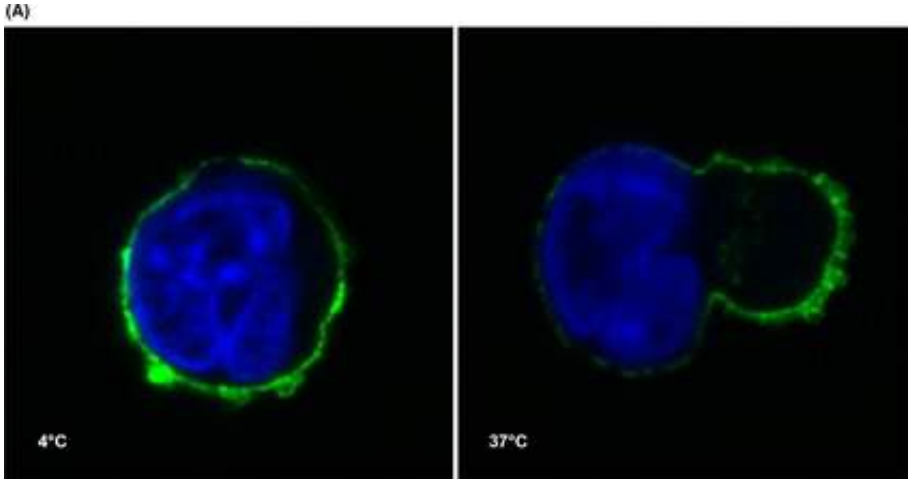
Leleu, Xavier, et al. *Ann Hematol. 2022*

Mechanisms of resistance to anti-CD38 mABs

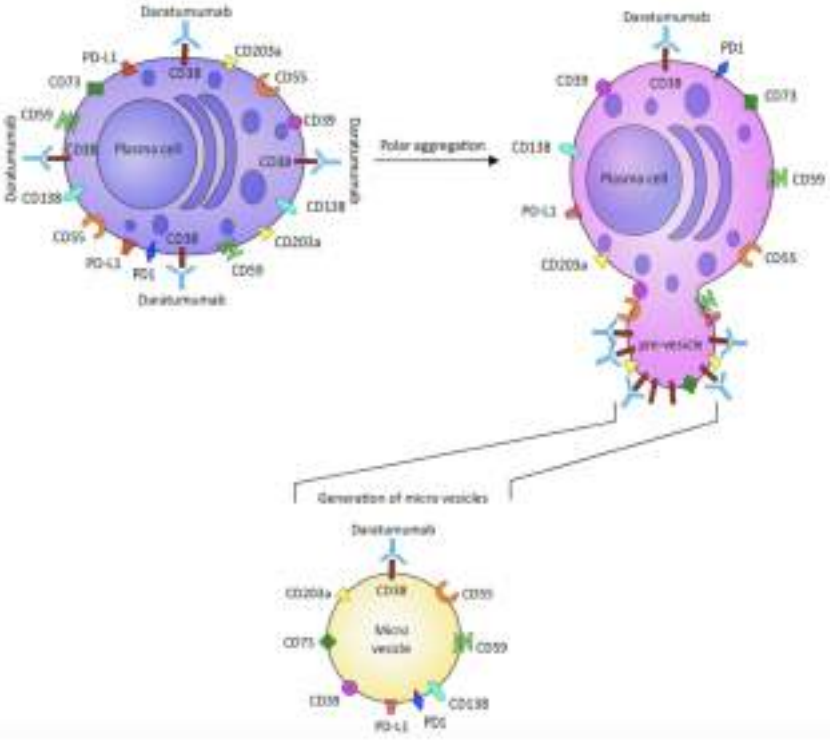


Modified form Saltarella I., Cells **2020**

Molecular surface dynamics of targeting CD38 by DARA in MM cells



Malavasi, F. et al. British journal of Haematology 2021



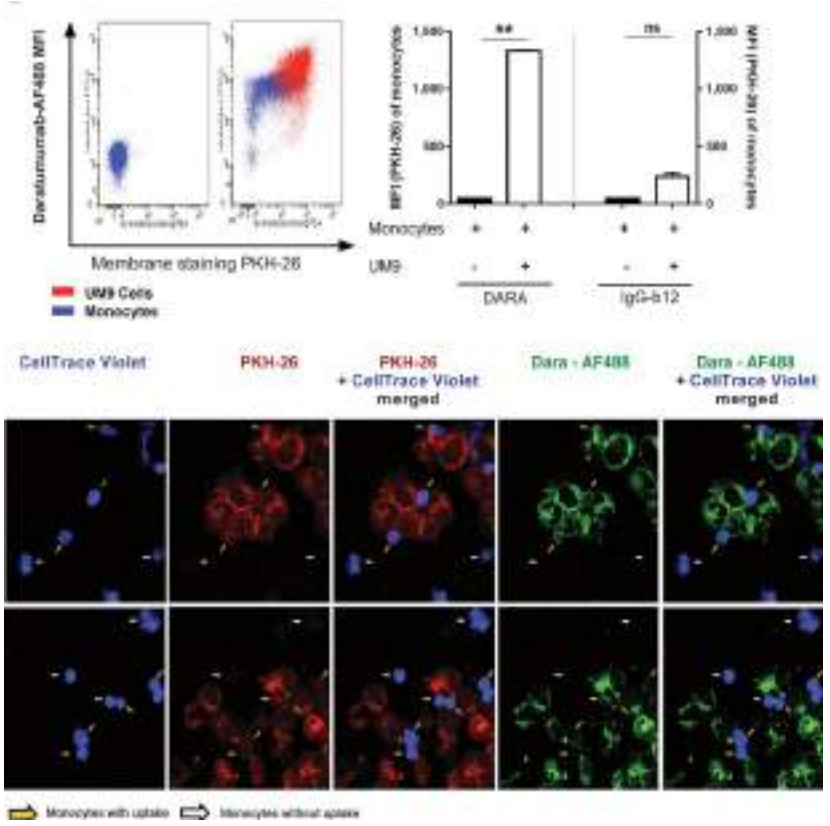
ISA induces internalization of CD38, but not its significant release from the surface of MM cells

Moreno L et al. Clin Cancer Res. 2019

Molecular surface dynamics of targeting CD38 by DARA in MM cells

Trogocytosis by monocytes and granulocytes:
- contributes to CD38 reduction,

- decrease of other surface proteins located nearby the CD38 antigen, including CD49f, CD56, CD54, and CD44

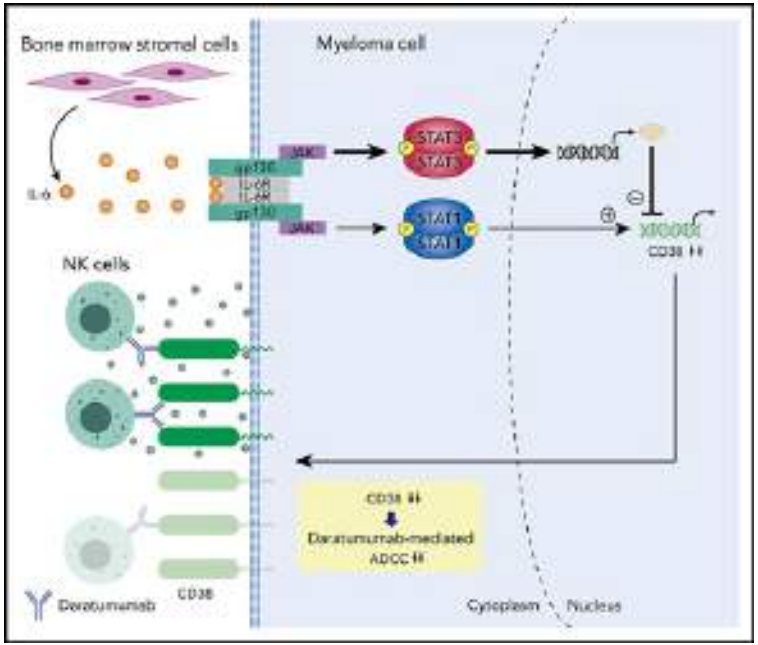


Krejcik, Clin Cancer Res (2017)

CD38 protein downregulation

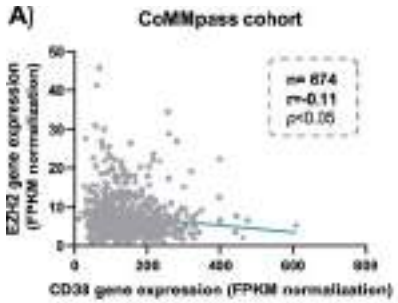
- Interleukin-6 (IL-6) plays a central role in MM cell proliferation and survival.
- CD38 expression on MM cells in the BM microenvironment is regulated by STAT3/IL6R (negatively).

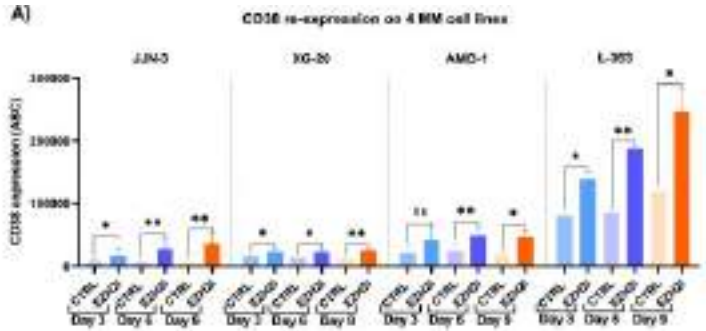
Ogiya D, et al. Blood. 2020
Kuroki, Br J Haematol, 2025

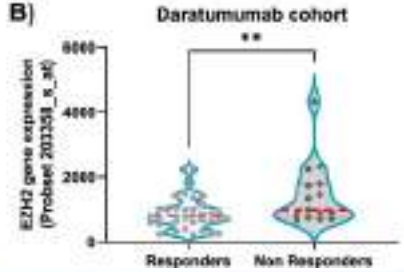


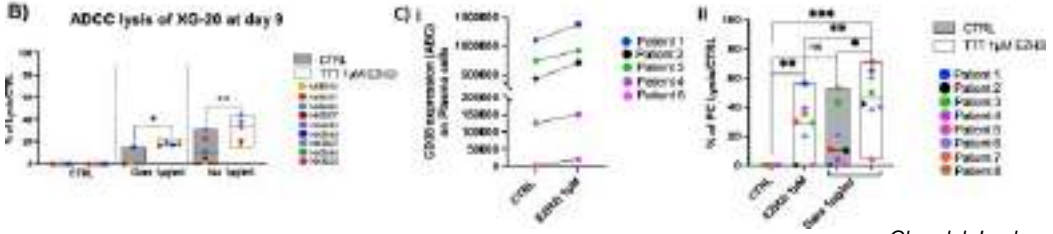
CD38 protein downregulation

- A significant negative correlation between CD38 and histone methyltransferase EZH2 expression
- Inhibition of EZH2 upregulates CD38 on surface and increase ADCC







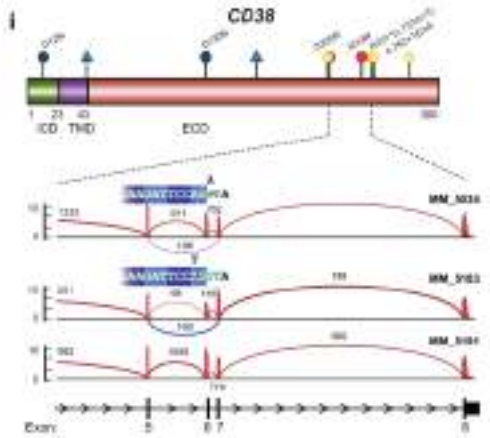


Chemlal, Leukemia 2023

Genomic antigen escape: point mutations and loss of CD38 gene

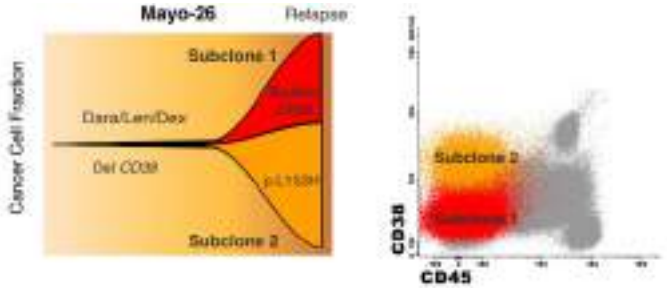
- In RRMM relative to NDMM point mutation induced in-frame exon skipping and removed most of the epitopes on CD38 that interact with DARA.
- These events may facilitate the evasion of MM cells from binding by DARA, while still retaining a major portion of the extracellular domain.

Vo, Josh N., et al. Nat Commun. 2022

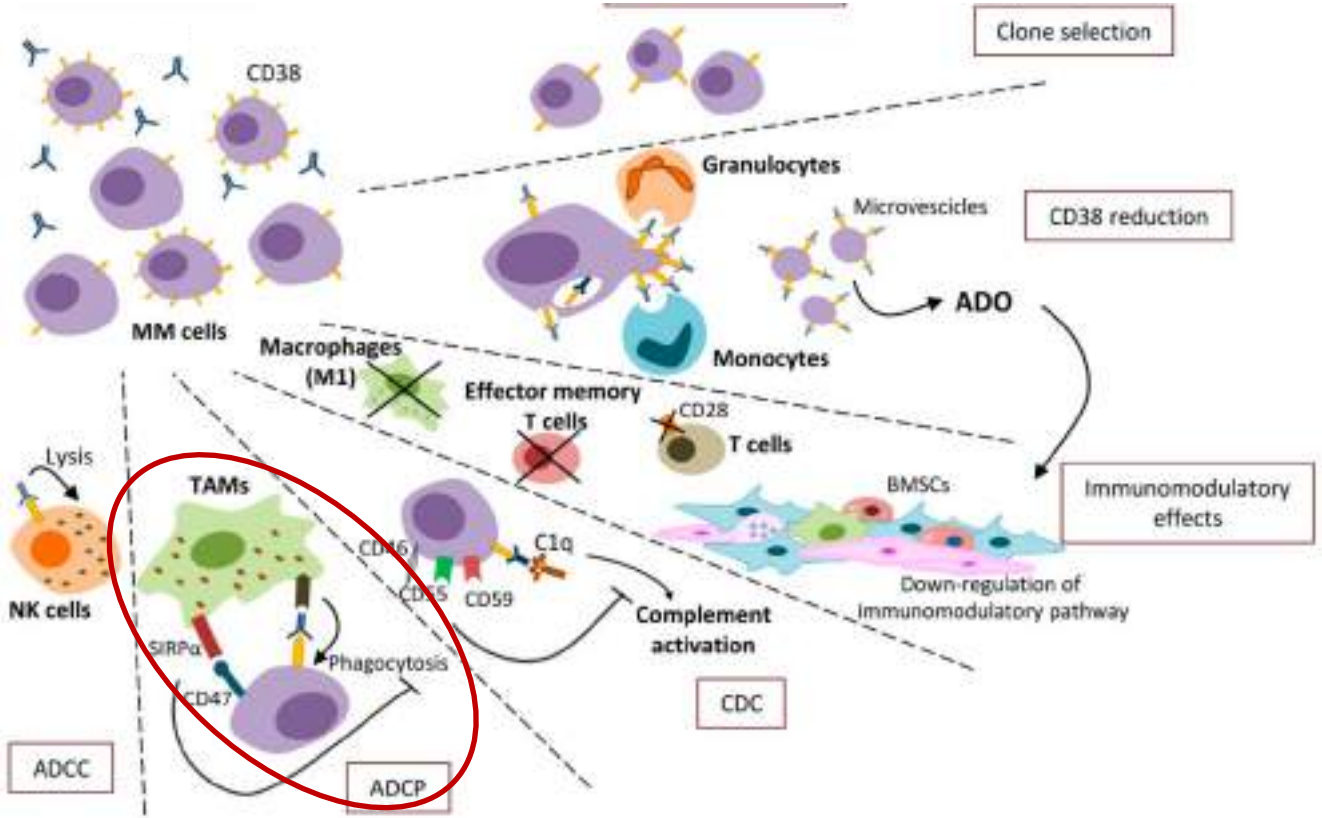


- The prevalence of monoallelic loss of *CD38* in newly diagnosed MM from MMRF CoMMpass was 7% but there were no cases of biallelic loss suggesting these **events are driven by treatment pressure**.
- Biallelic inactivation of *CD38* at anti-CD38 MoAb relapse was 6%.

Diamond B., Blood (2024) 144 (Supplement 1)



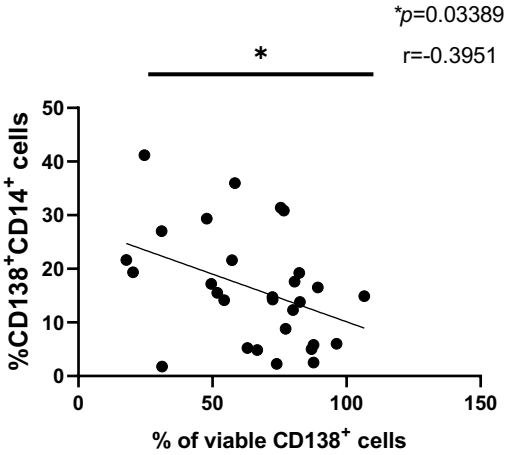
Mechanisms of resistance to anti-CD38 mABs

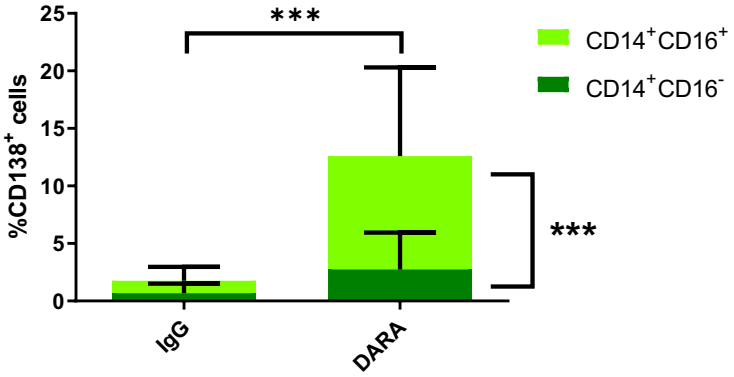


Modified form Saltarella I., Cells **2020**

Resistance to ADCP: «don’eat me» signal

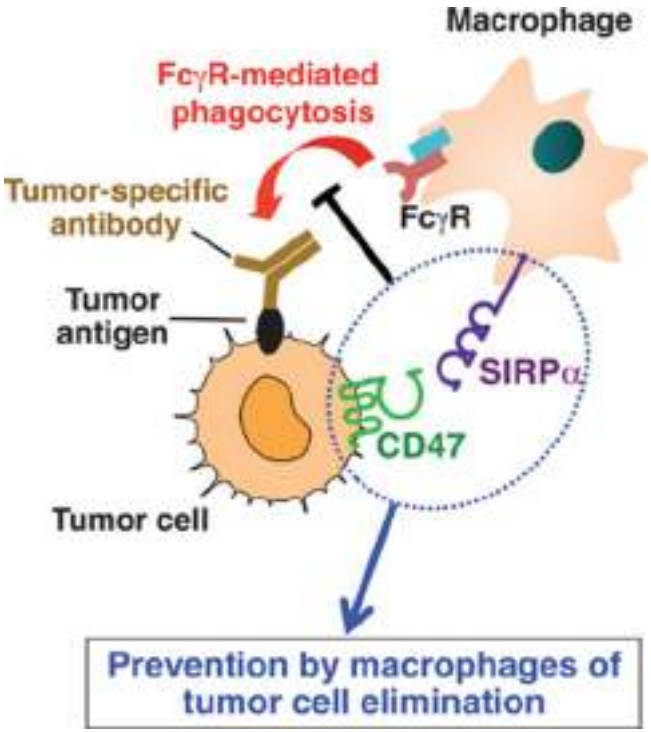
- The CD138+CD14+ double population is correlated with the efficacy of DARA and with the CD14:CD138 ratio;
- The monocyte engaged in the CD138+CD14+ population are CD16+

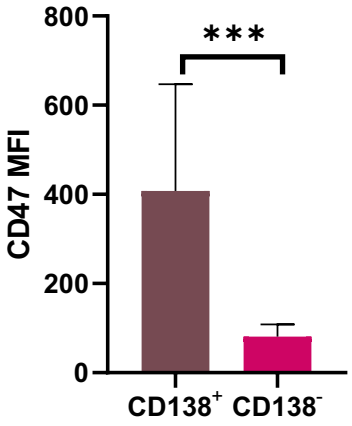


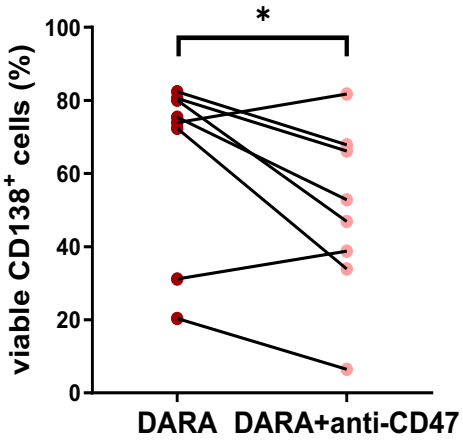


Storti P. et al, British Journal of Haematology 2019

Resistance to ADCP: «don’eat me» signal



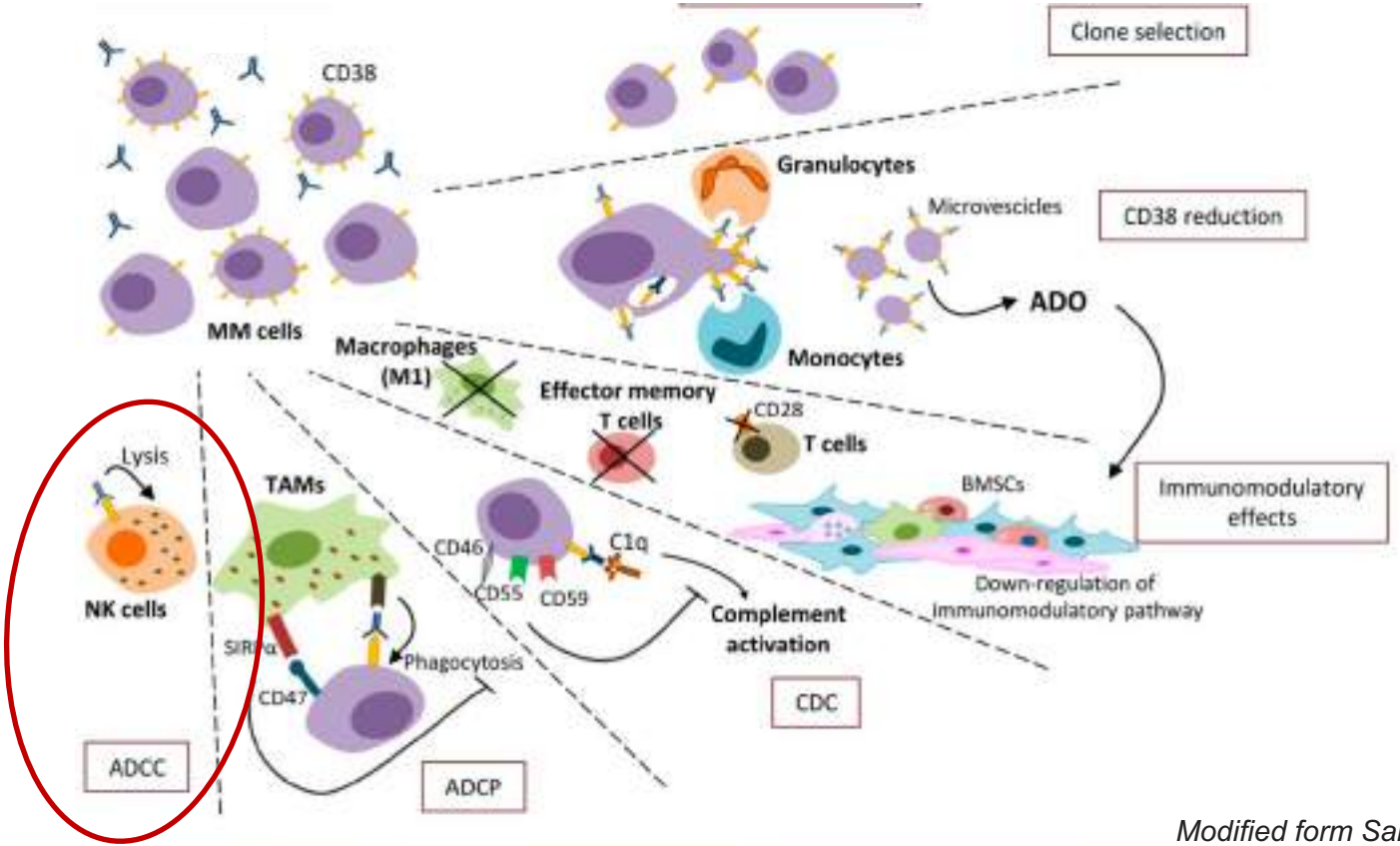




CD47 neutralization increases the anti-MM activity of DARA

Storti P. et al, British Journal of Haematology 2019

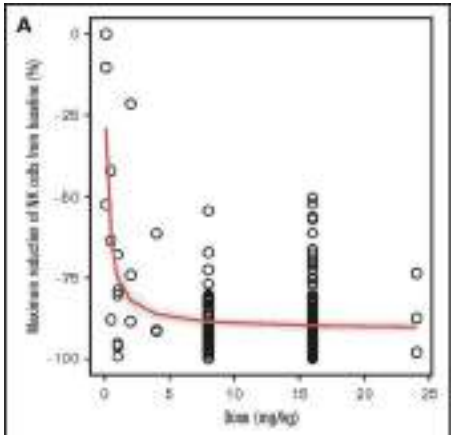
Mechanisms of resistance to anti-CD38 mABs

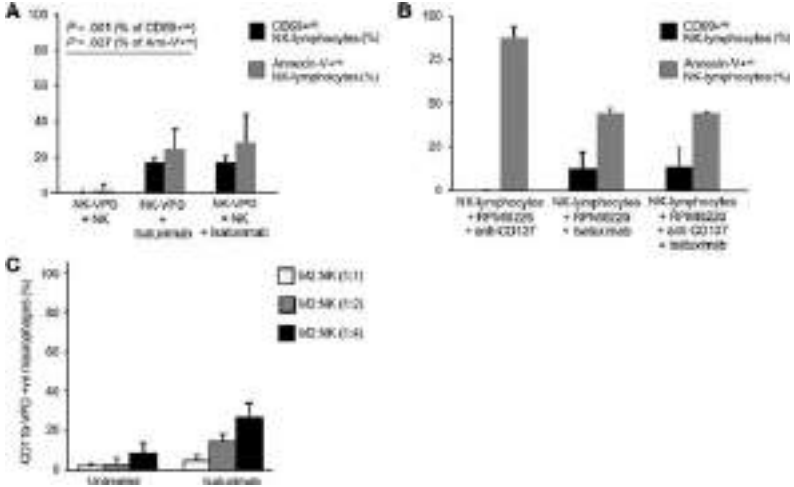


Modified form Saltarella I., Cells **2020**

Resistance to ADCC

NK cells also express CD38, a rapid reduction in NK cells in both peripheral blood and bone marrow is observed after DARA and ISA treatment in both responders and non-responders patients



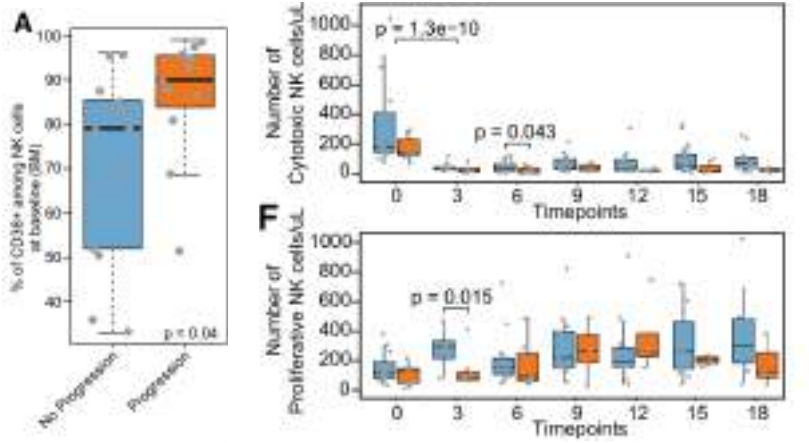


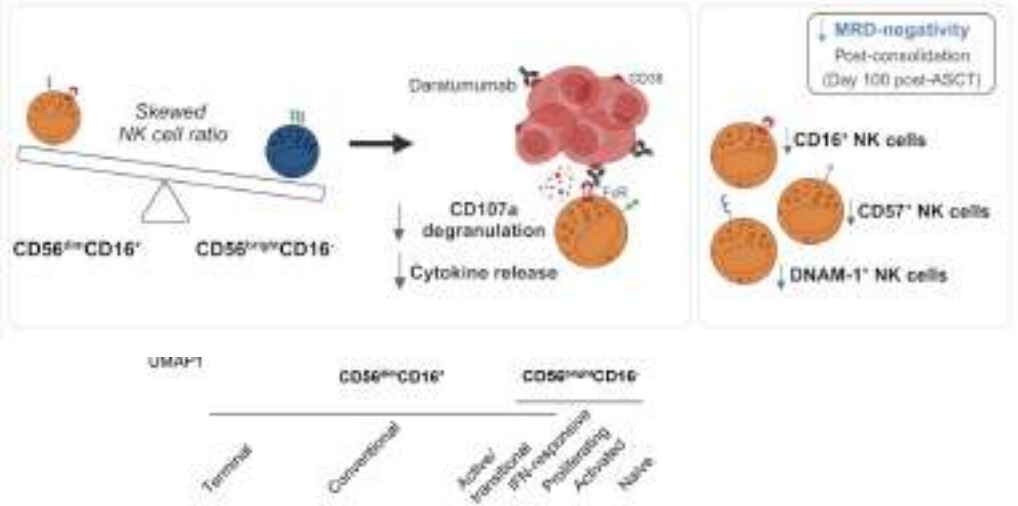
Casneuf, Blood Adv (2017)

Moreno, Clinical Cancer Res 2019

Resistance to ADCC: NK subset dynamics

- The expansion of proliferative NK cells was significantly higher in durable responders compared to progressors
- A low proportion of CD16+ BM NK cells was associated with reduced likelihood of achieving MRD-negativity upon treatment with D-VTd



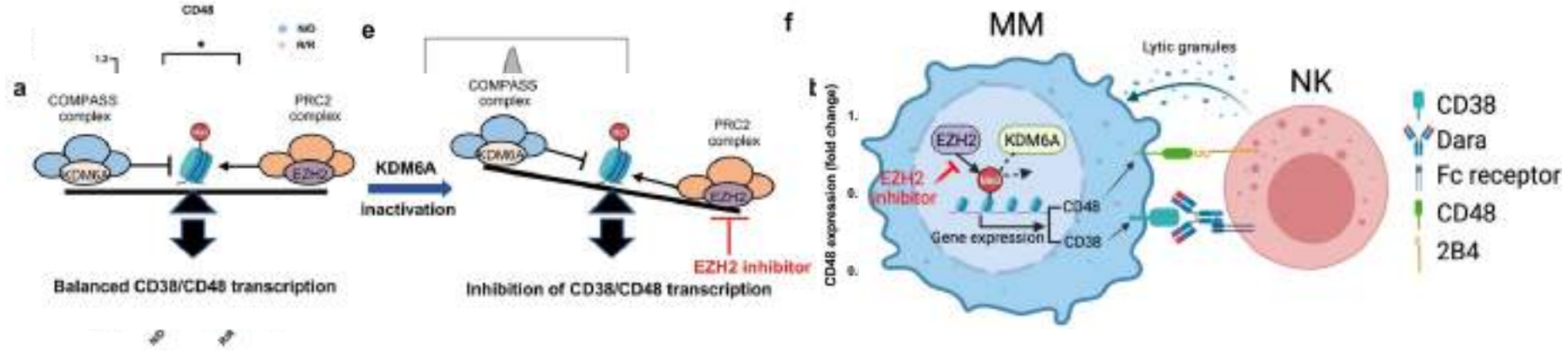


Zicchedu, Blood Cancer Journal, 2024

Korst, Blood 2025

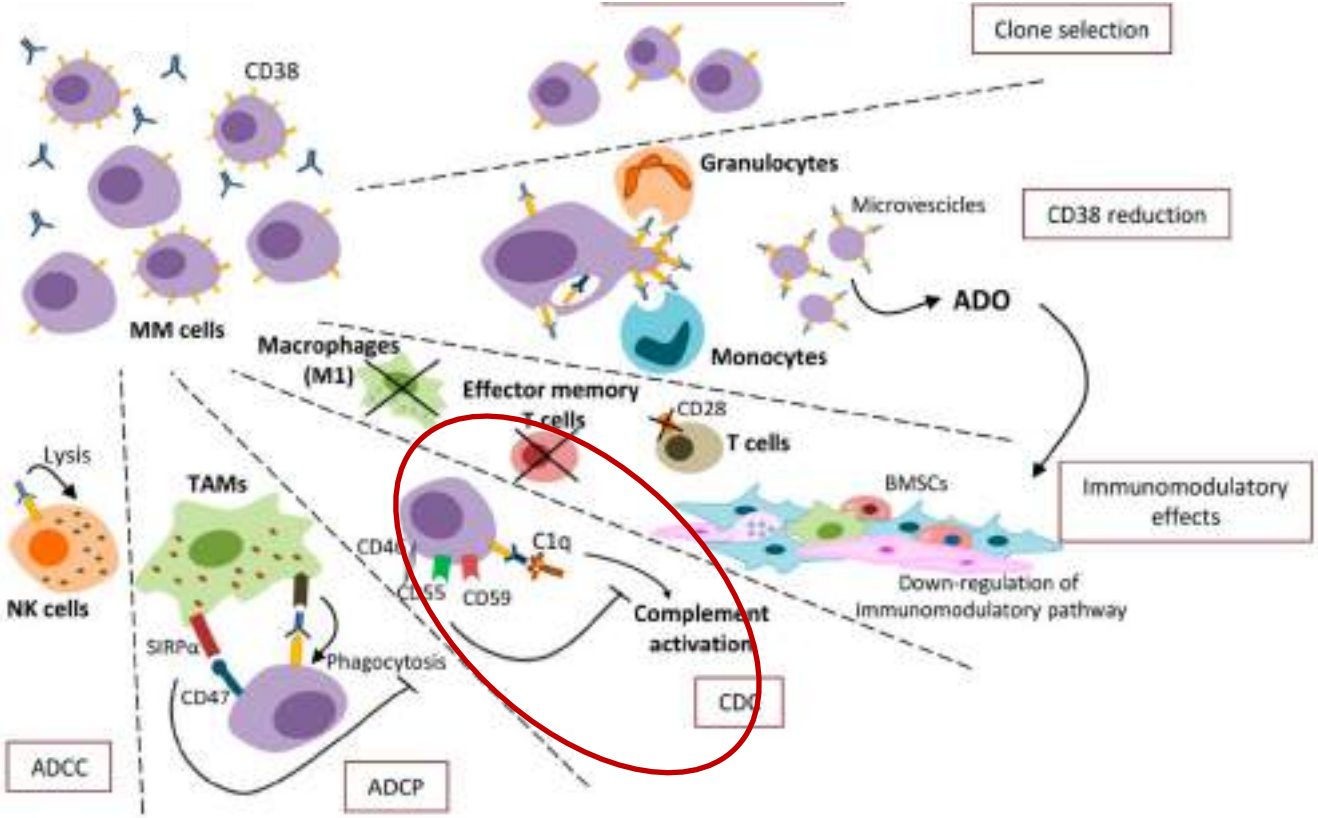
Resistance to ADCC: CD48 downregulation

- CD48 expression levels in Dara-resistant patient MM samples are lower than in the newly diagnosed MM
- The loss or inactivation of *KDM6A* increased the level of H3K27me3, resulting in the downregulation of both CD38 and CD48 expression, which led to reduced ADCC.



Liu J., Nature Communications 2024

Mechanisms of resistance to anti-CD38 mABs

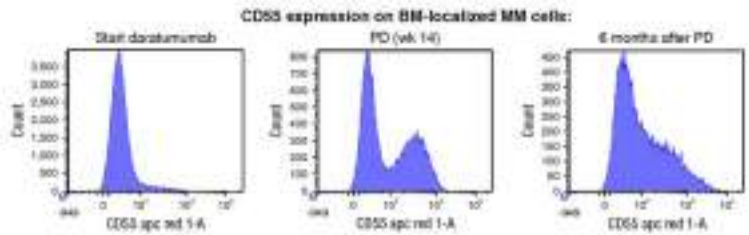


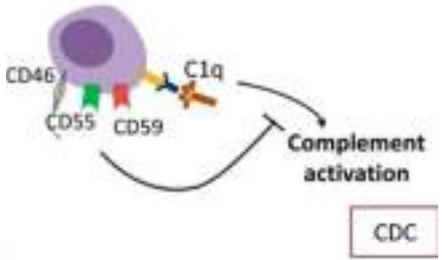
Modified form Saltarella I., Cells **2020**

CDC inhibition

Increased expression of CD55 and CD59 prevents CDC[2]

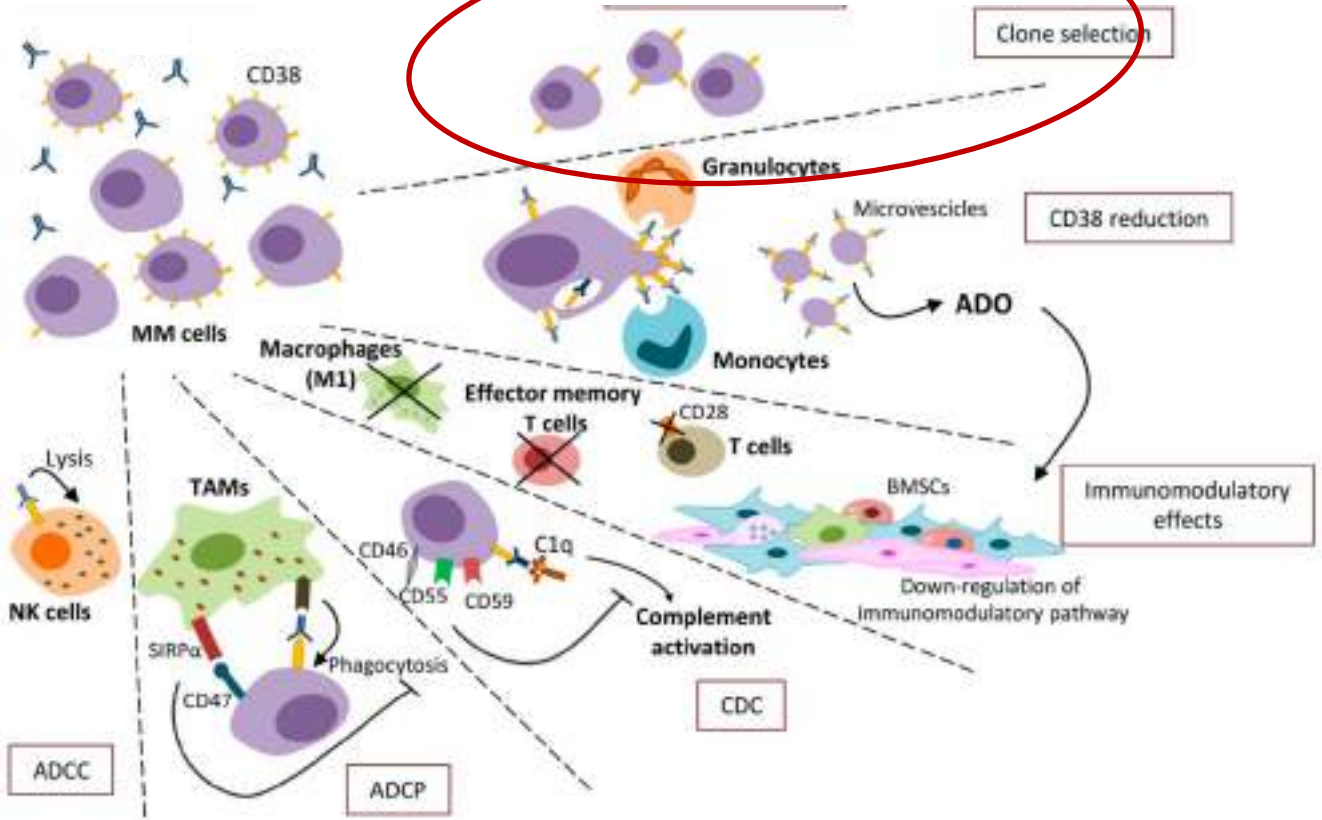
Increased expression of CD55 and CD59 is seen on MM cells during disease progression on Dara therapy[3,4]





[1]Bisht K, et al. *Expert Rev Hematol.* 2021
[2]Saltarella I, et al. *Cells.* 2020
[3]Nijhof IS, et al. *Blood.* 2016
[4]Zhu C, et al. *Front Immunol*. 2020

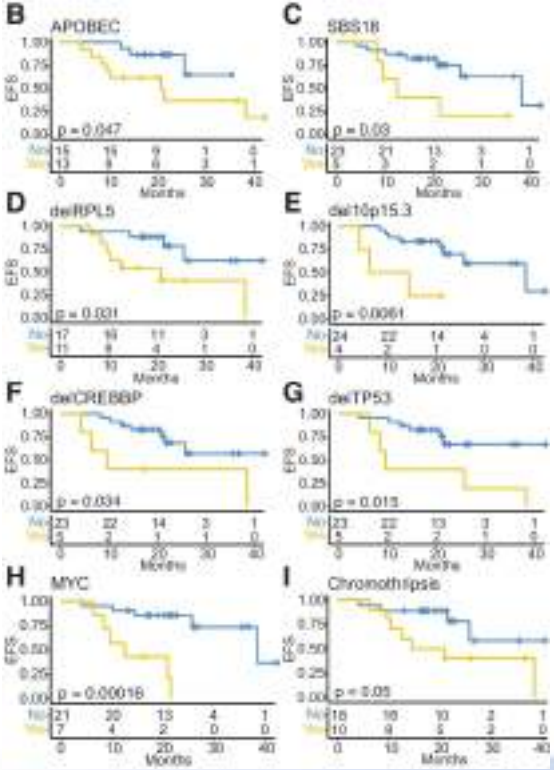
Mechanisms of resistance to anti-CD38 mABs



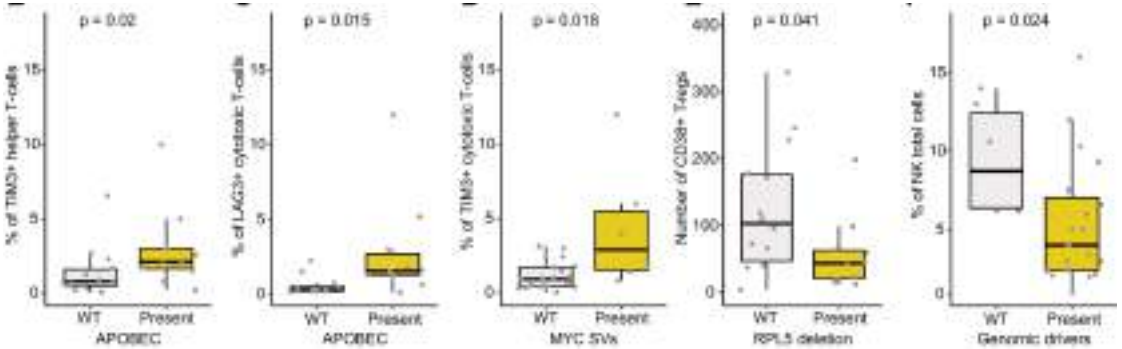
Modified form Saltarella I., Cells **2020**

Genomic determinats of resistance to anti-CD38 mAbs

Early progression after dara-VTd: RPL5 loss, APOBEC mutagenesis, or gain of function/ structural variants involving MYC or chromothripsis.



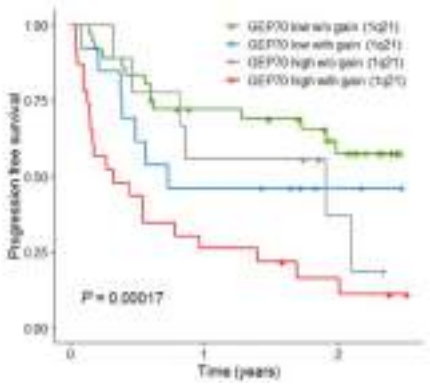
Patients with high APOBEC mutational activity were characterized by an enrichment of exhausted T cells and reduced baseline number of NK cells was observed in all genomic features associated with progression.

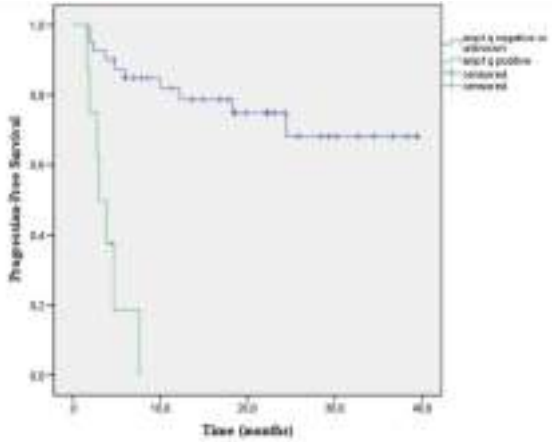


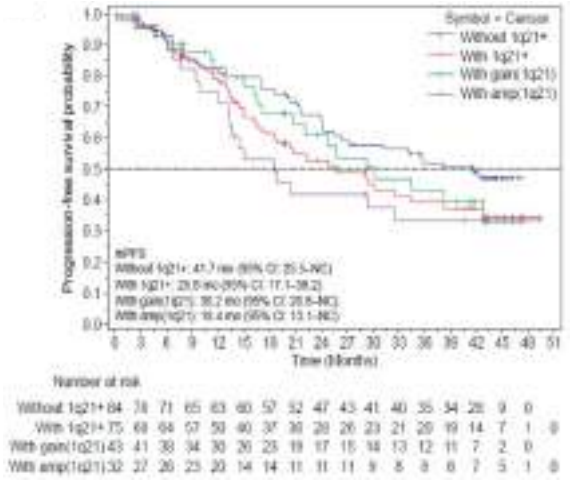
Zicchedu, Blood Cancer Journal, 2024

Genomic determinats of resistance to anti-CD38 mAbs

- The presence of a high-risk GEP70 score significantly impacted OS negatively with daratumumab treatment
- The presence of gain 1q21 at initial presentation negatively impacted PFS and OS with daratumumab treatment.
- A PFS benefit was observed with Isa-Kd vs Kd in all subgroups evaluated also 1q21+







Mohan M, et al, *Br J Haematol. 2020*

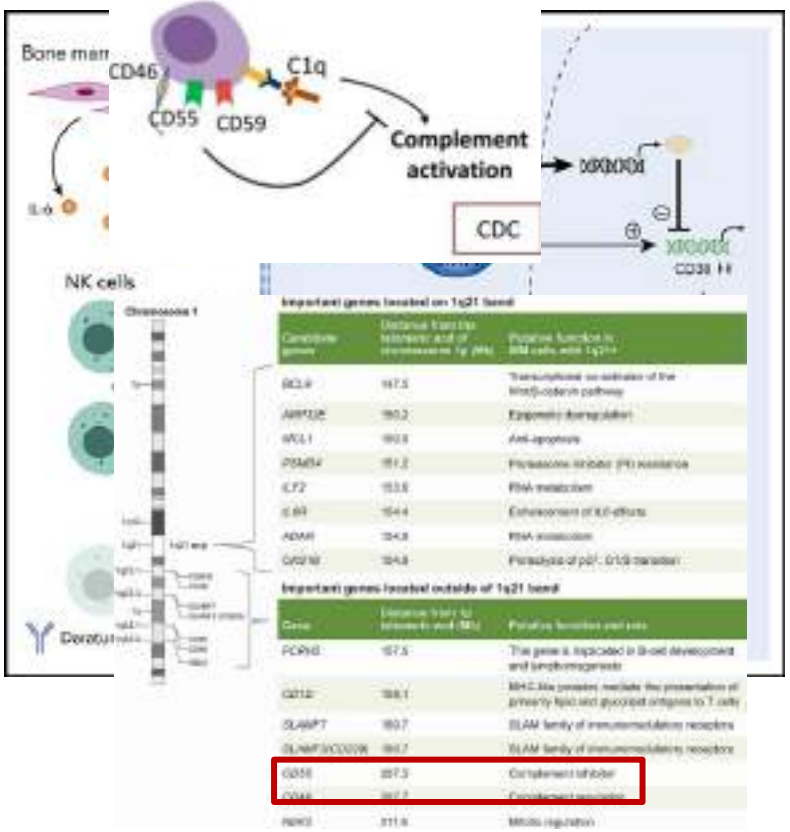
Barbieri E, et al. *Ann Hematol. 2022*

Facon T, et al. Hematol Oncol. 2024

1q21+ and resistance to anti-CD38 mAbs

IL6 receptor (IL-6R) is located on chromosome 1q21 and IL-6 plays a central role in MM cell proliferation.

Patients with 1q+ overexpress CD55 and thus may have high resistance to drugs that rely on CDC activation.



[1]Bisht K, et al. *Expert Rev Hematol.* 2021
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Ogiya D, et al. Blood. 2020 Nov 12;136(20):2334-2345

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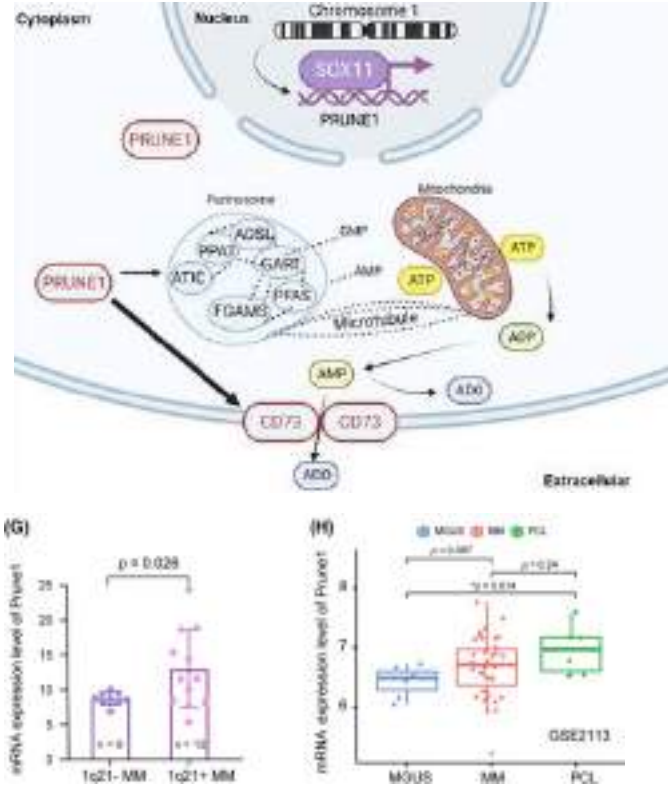
PRUNE1 CN is adversely associated with the survival of MM patients with 1q21+.

CD73 is a downstream target of PRUNE1 in MM cells with 1q21+, which also explains the high heterogeneity of CD73 expression in patients with MM and higher ADO production



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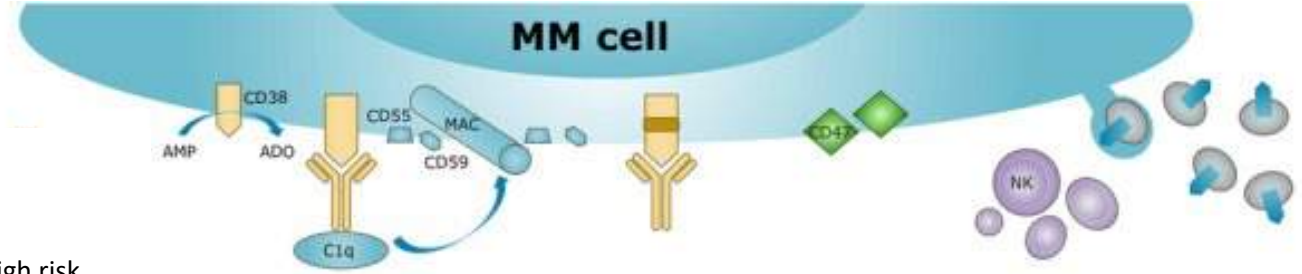
[4]Zhu C, et al. *Front Immunol*. 2020

Ogiya D, et al. Blood. 2020 Nov 12;136(20):2334-2345

Xu J,et al. *Br J Haematol.* 2023



Resistance mechanism	High risk genomic alterations	Downregulation of CD38	CD55 and CD59 overexpression inhibits CDC	Mutated or splice variants of CD38	CD47 overexpression inhibits ADCP	TIM-3⁺ and HLA-DR⁺ exhausted CD16⁻ NK-cells	Release of microvesicles containing CD38

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Thanks for the attention

Hematology Laboratory
Department of Medicine and Surgery
University of Parma

Multiple myeloma and monoclonal gammopathies program
Hematology AOU Parma

Prof. Nicola Giuliani

Denise Toscani
Nicolas Thomas Iannozzi
Rosanna Vescovini
Vincenzo Raimondi
Oxana Lungu
Gabriella Sammarelli
Giannalisa Todaro

Benedetta Dalla Palma
Federica Librale
Matteo Scita
Matia Bernardi
Stefania Ricci

UNIVERSITÀ DI PARMA

AIRC

MMRF MULTIPLE MYELOMA Research Foundation

ParmAIL