Short overview, including vector technology, types of antigens

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Antigens are at the centre of immuno-oncology approaches

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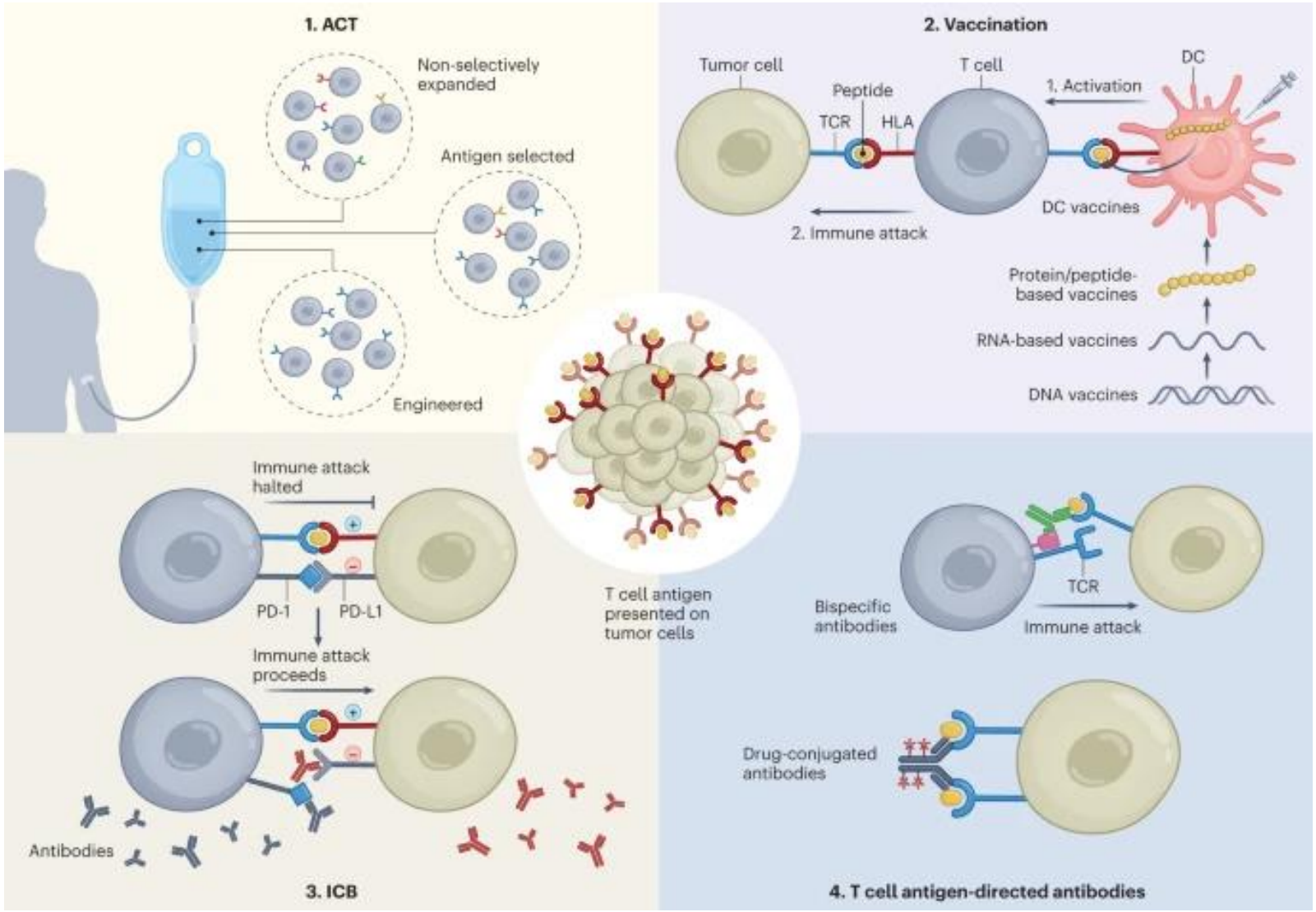
ACT

VACCINES

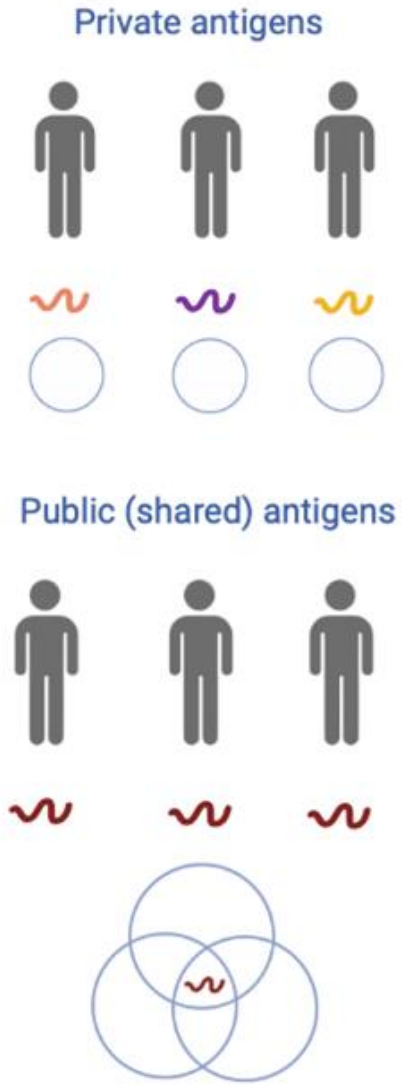


ICB

ADCs/Bi-specifics

Peri et al. Nature Cancer 2023

Outline



1. Comparison s of shared versus private antigens:

advantages, challenges, therapy targeting

2. Multi-omic approaches to investigate antigens driving immune

responses in patients

Shared Antigens: Examples

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Differentiation antigens	MelanA, gp110, tyrosinase
Aberrantly expressed tumor-associated antigens	Her2, Muc-1
Germline antigens	MAGE family, NY-ESO-1
Stemness antigens	SOX2, OCT4
Viral oncoproteins	HPV E6
Recurrent somatic mutations	B-Raf V600E. KRAS G12D

Shared Antigens: Advantages

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- Common across patients and even cancer types
- Established antigen profiles with therapeutic precedents (e.g. HER2)
- Scalability: streamlined production for T-cell therapies or vaccines
- Logistical simplicity relevant for population approaches, e.g. cancer prevention
- Shared shared antigens and TCRs are particularly relevant for tumour types with low neoantigen burden (low TMB)

Shared Antigens: Challenges

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- Off-target effects, healthy tissue toxicity
- Immune Tolerance (low “foreignness” except for viral antigens and recurrent neoantigens)
- Tumour heterogeneity: variable expression within a tumour; could lead to resistance
- Targeting a single antigen poses a selective pressure for resistance to emerge

Shared Antigens: therapy success

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TCR therapies

- NY-ESO-1 TCR-T
- Tebentafusp gp100-specific TCR fused to an anti-CD3 effector molecule
- TCR-engineered T cells targeting E7

Vaccines

Evidence of efficacy with TAAs targeting (RNA, DNA, peptide) +/- ICI

PLACEHOLDER OVERVIEW OF VACCINE TRIALS

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Unique Antigens - Neoantigens

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- **Non-synonymous mutations**
- Insertions/deletions
- Retained introns
- Gene fusions
- Not present in normal tissues (CAVEAT: increasing evidence of normal tissue mosaicism including for cancer driver mutations)

Unique Antigens: Advantages

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- Personalized approach
- Specificity - less of target effects and greater potential for combination therapy)
- Increased immunogenicity through increased ”Foreignness”
- Potential for targeting **multiple** private neoantigens mitigating risk of resistance due to antigen loss or HLA loss

Unique Antigens: Challenges (1)

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Neoantigen discovery is involved

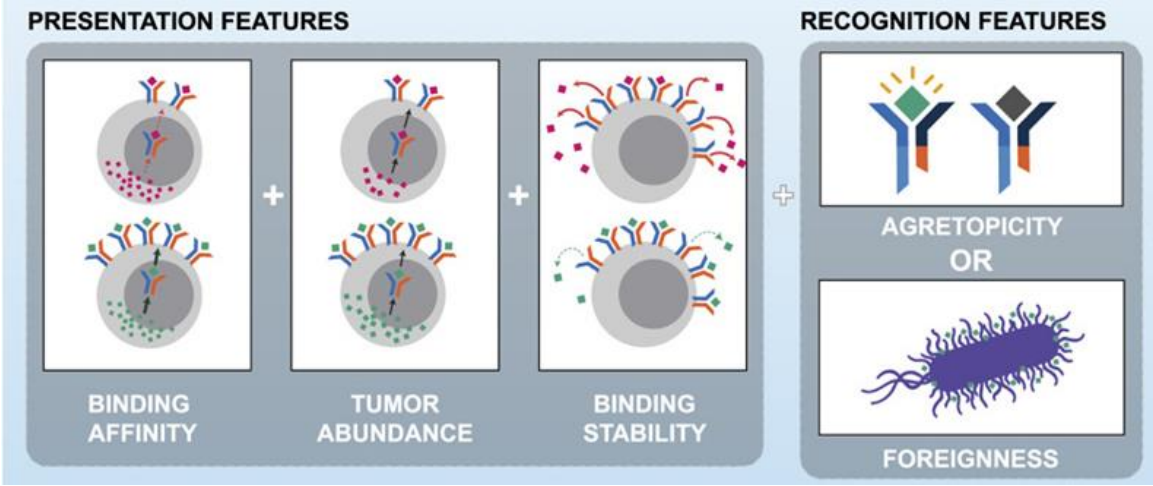
Steps needed to identify neoantigens

DNA (WES) sequencing

1. HLA typing
2. Somatic mutation calling
3. Assessment of mutation clonality (SCNAs)
4. Neoantigen prediction and prioritisation
    - Binding affinity or elution probability
    - Binding stability
    - “Non-self” score
    - Distance to WT peptide
5. LOH of HLA alleles

RNA sequencing

1. Assessment of abundance
2. Downregulation of HLA alleles



Neoantigen calling is unreliable!
Less than 1% of mutations are predicted as strong neoantigens and only 6% of these are validated as immunogenic

(Wells et al., 2020, Cell)

Unique Antigens: Challenges (2)

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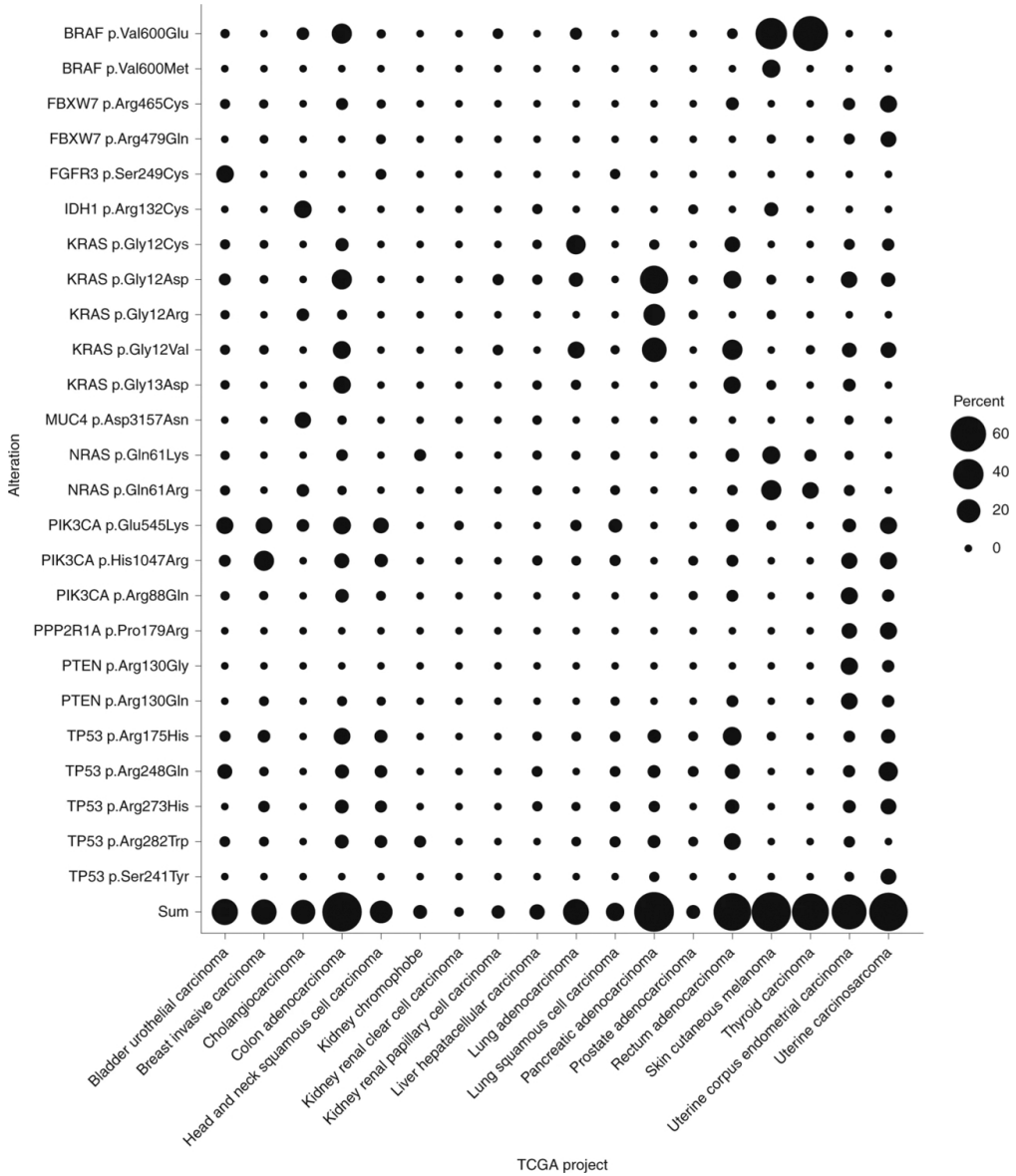
- High TMB tumours are the primary focus (given TMB correlation with neoantigens)
- Neoantigens may be subclonal due to intratumour hetreogeniety – easily immunedited
- High cost and complexity of development
- Longer lead time cf off-the self product

Recurrent neoantigens may overcome some of these challenges?

Recurrent somatic mutations are potential public neoantigens



Could be off the shelf products

Target neoantigens more likely to be clonal as drivers

BUT

Require frequent HLA type

Higher selective pressure as targeting fewer neoantigens (just one)

Frequency of recurrent mutations across cancer types. Pearlman, Nature Cancer 2022

Unique Antigens: therapy success

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Cell Therapy

- lifileucel for melanoma, approved
  - Unselected TILs
- Neoantigen reactive TILs
  - NCT03412877, Achilles Therapeutics

Vaccines

- Clinical benefit in combination with PD1
- Up to 20 neoepitopes selected per patient
- Can improve subdominant neoantigen response (Burger et al., 2021, Cell)

Vaccines against driver mutations

- KRAS targeting in NSCLC with ICI

refs:

What are tumour cells seeing in the TIL product- Melanoma

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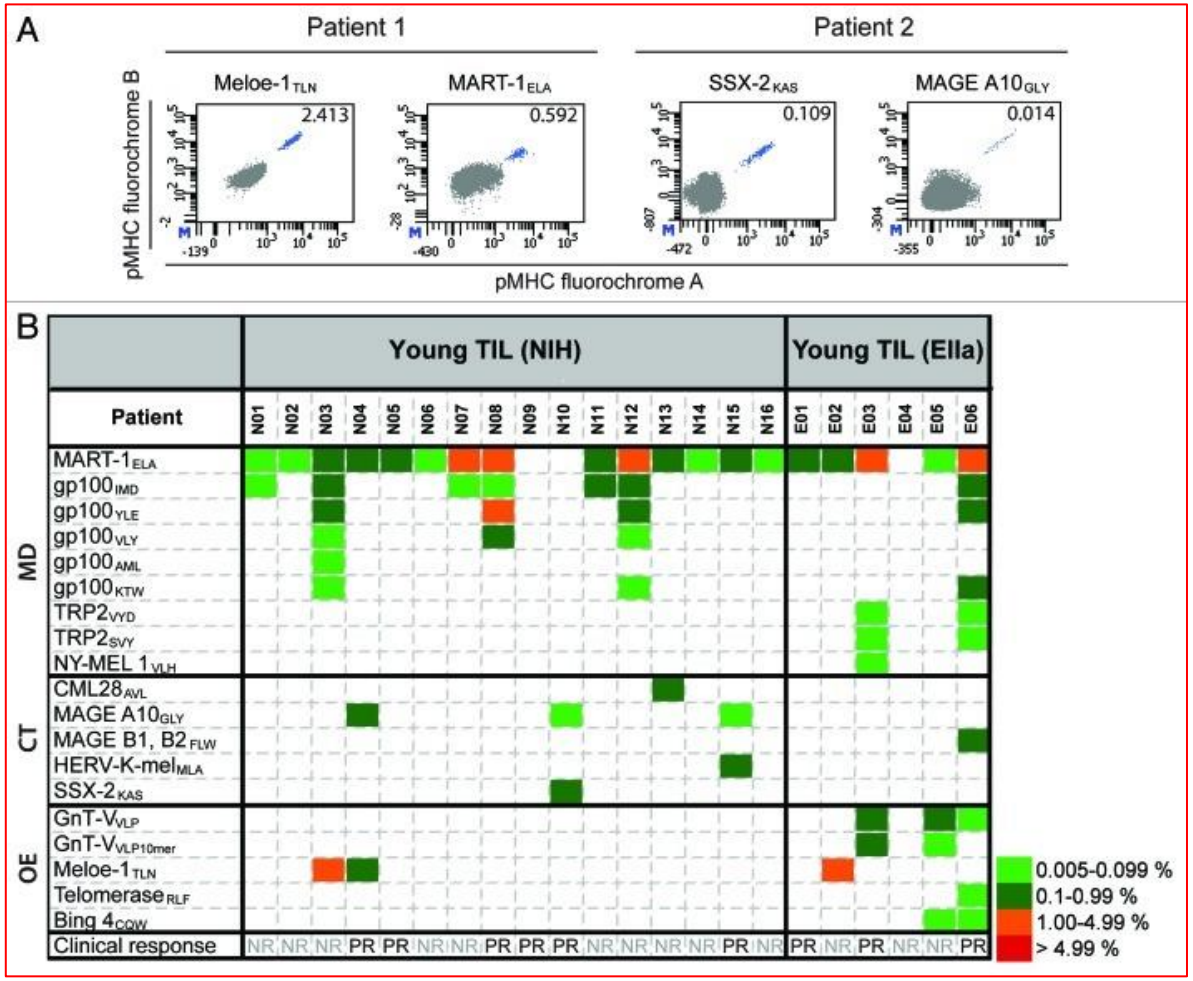
TIL product: Melanoma-associated antigens



Kvistborg Oncoimmunol 2012

TIL product: Melanoma Neoantigens

	Gene mutation	Frequency (% multimer⁺CD 8⁺ T cells of total CD8⁺ T cells)	Restriction element
Patient 3	RBM12 $_{S>L}$	29.00	HLA-B*51:01
	VARS $_{T>M}$	0.096	HLA-A*25:01
	MYLK $_{G>V}$	1.90	HLA-A*25:01
	LRP3 $_{T>S}$	0.039	HLA-A*25:01
	WDR1 $_{N>K}$	0.50	HLA-A*68:01
Patient 4	TTC37 $_{A>V}$	1.14	HLA-A*01:01
	ENTPD4 $_{P>L}$	3.35	HLA-A*01:01
	MAB21L1 $_{V>M}$	0.007	HLA-B*08:01
Patient 8	RAD51AP1-002 $_{S>F}$	0.195	HLA-A*03:01

Van Der Berg JITC 2020

Another factor to consider: is high immunogenicity too much of a good thing?

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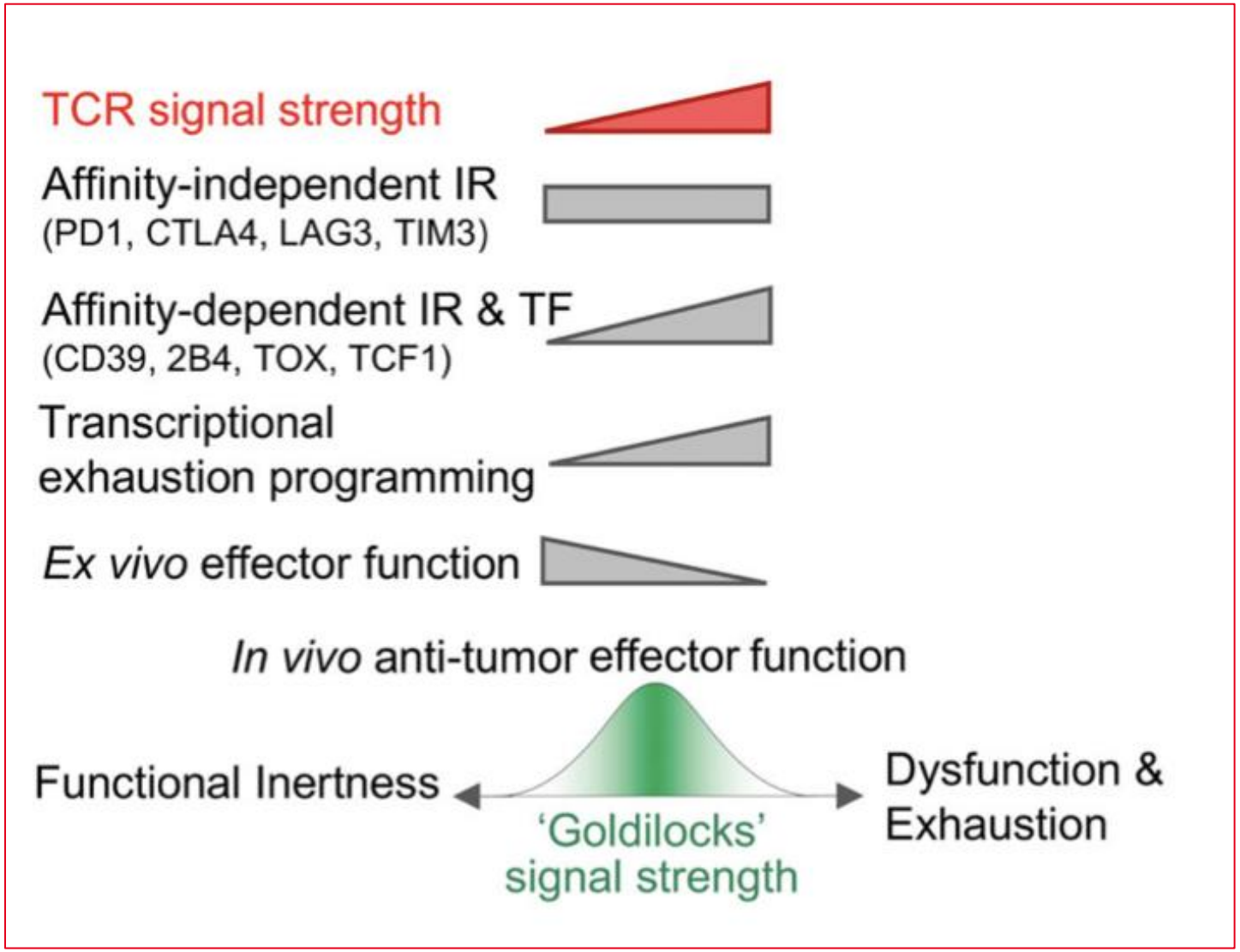
Low immunogenicity Antigens

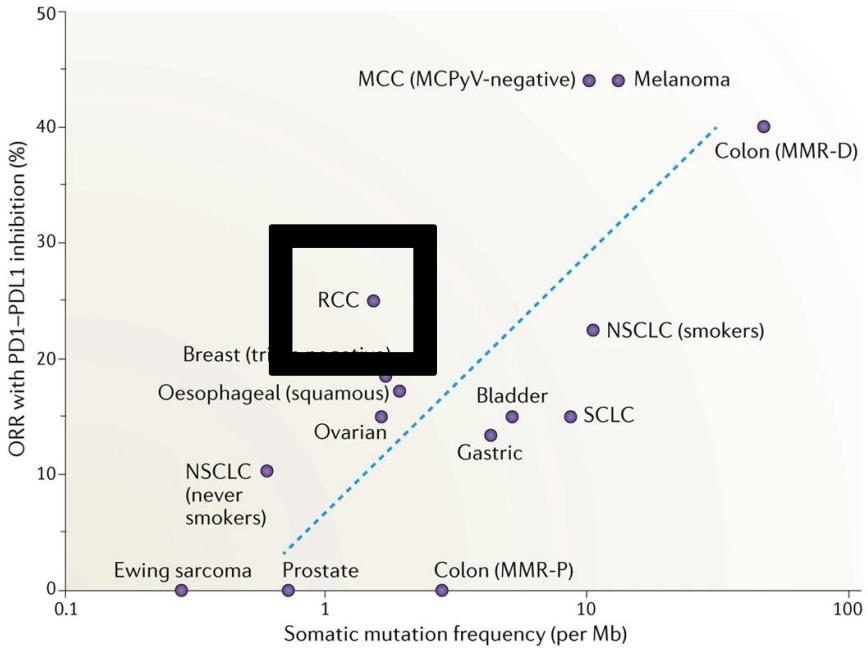


High immunogenicity Antigens

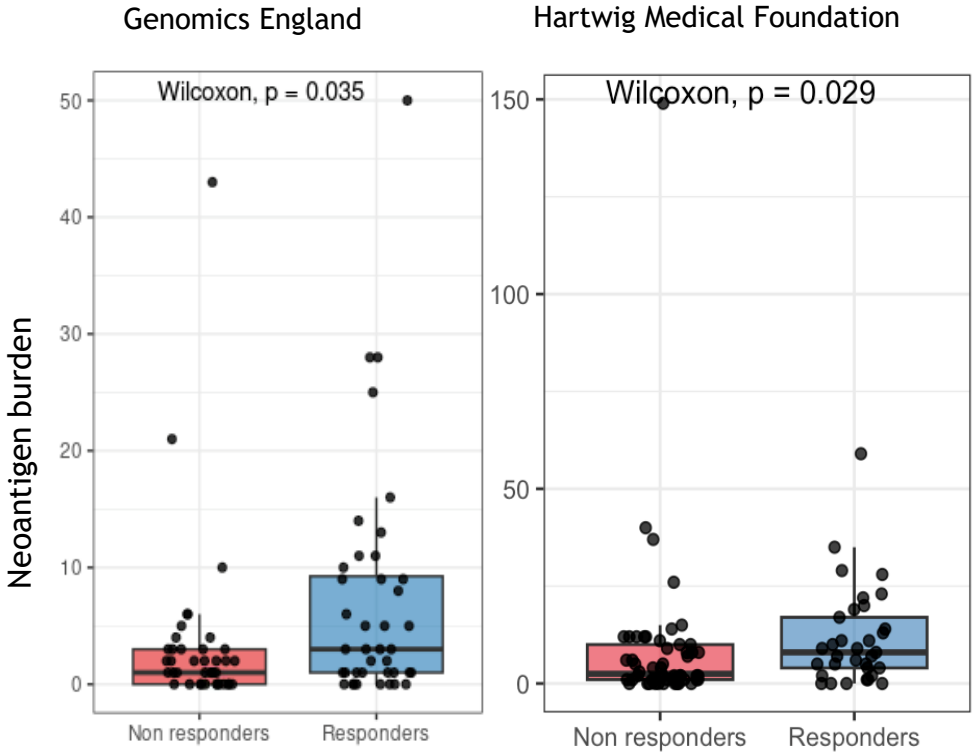
Shakiba et al, JEM 2021

In favour of neoantigens

- TMB and ICI response relationship
- predicted neoantigen burden is higher in tumours that respond to ICI
- primed T cells reinvigorated by ICI <u>often</u> recognise **neoantigens**



(Yarchoan et al 2017, Nat Rev Can)



Unpublished data - Irene Lobon

Frameshift mutations are enriched in RCC but fail to explain ICI response

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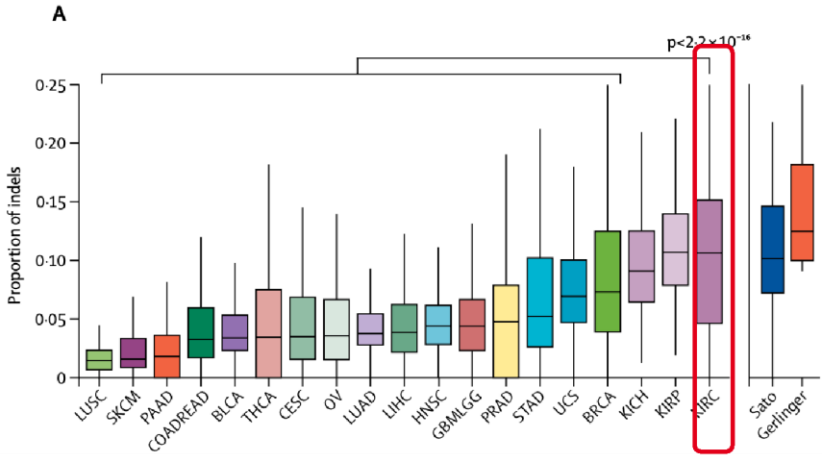
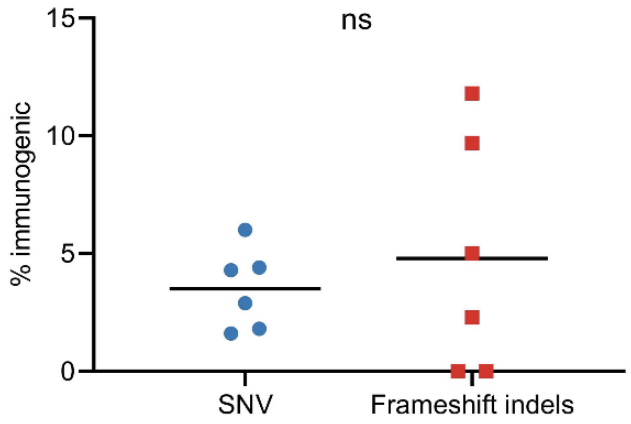
Proportions of indels in ccRCC are higher than in other solid cancers



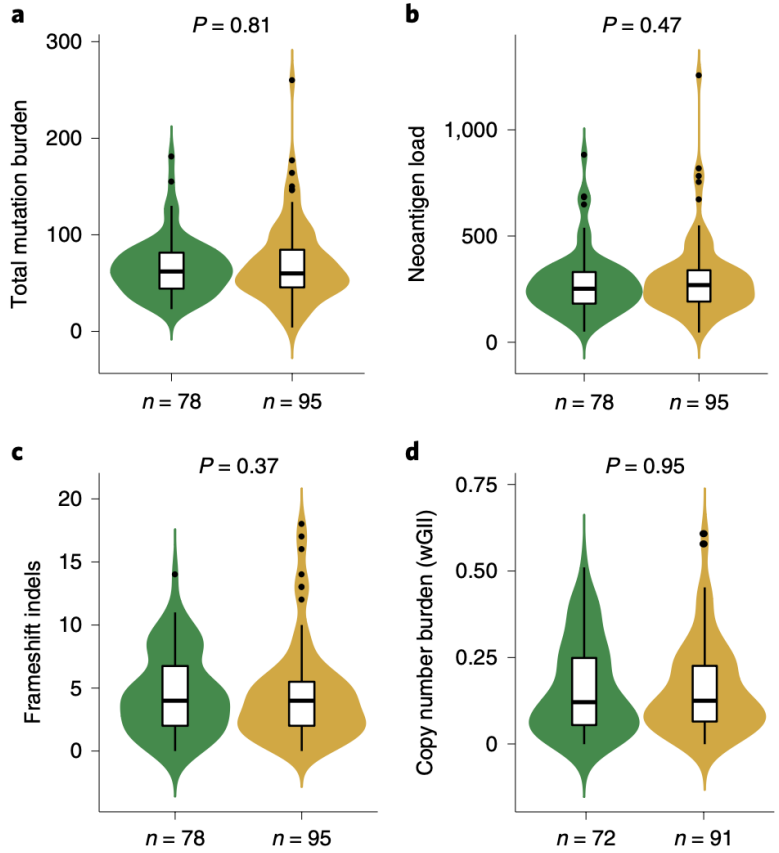
Turajlic and Litchfield, 2017

FS-indels and SNVs specific CD8 TILs are found in ccRCC patients



Hansen et al, Front. Immun. 2020

TMB and FS-indels do not predict CPI clinical benefit



Braun et al, Nat. Medicine 2020

Are they recognizing an unconventional antigen?

Unconventional antigens – potentially shared but underexplored

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- Alternative splicing
- Post-translational modifications
- **Expressed endogenous retroelements (ERVs)**
- Epigenetically silenced in normal somatic tissue, but de-repression and transcription can occur in disease states including cancer
- Non-coding epitopes, could account for up >80% of tumor specific antigens *Laumont et al. 2018 STM*
- Bacterial antigens

Immunogenicity, Tolerance, Heterogeneity ?

Conclusions: targeting shared versus private antigens

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- Pros and cons with both approaches
- Depends on TMB, HLA restriction

- Ideally target both and multiple as likely common mechanisms of resistance
- 1. antigen loss/downregulation
- 2. loss of antigen presenting machinery

- Ares of unmet need:

1. induction of memory/long-term responses
2. Investigation of unconventional antigens