Impact of checkpoint inhibitor therapy on somatic mutations: Tumors become more stealth

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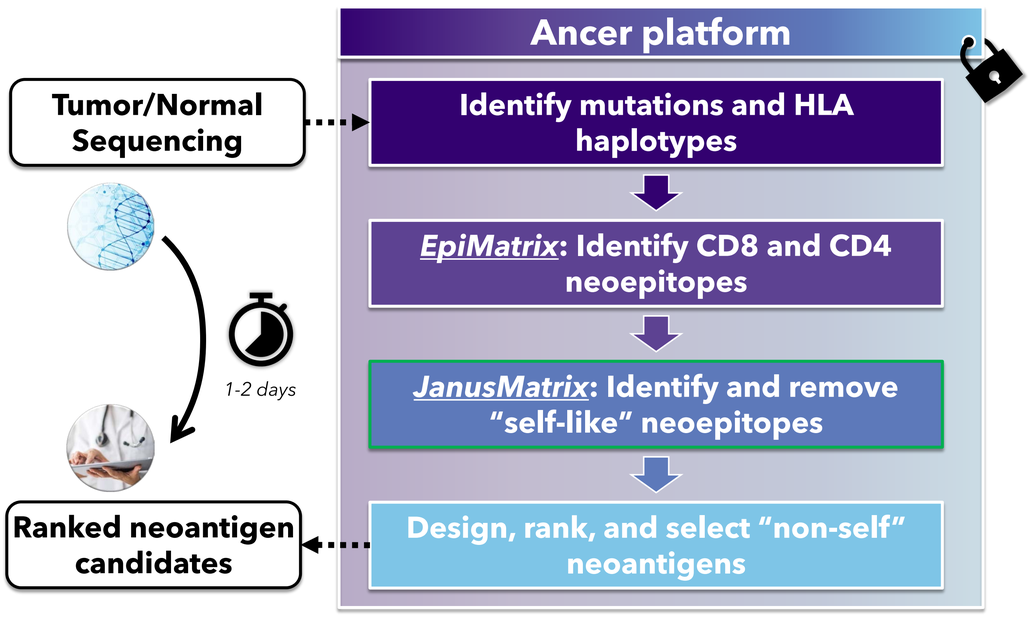
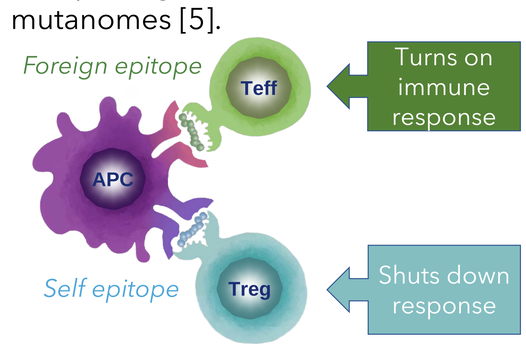
Overview

- **Hypothesis**: Tumor clones surviving checkpoint inhibition therapy harbor mutations more prone to immune avoidance.
- **Approach**: Tumors from melanoma patients collected **before and after** nivolumab immunotherapy (n=41) were analyzed with **Ancer, an advanced neoepitope screening platform** that combines proprietary machine learning-based **CD8 and CD4** epitope mapping tools with removal of **inhibitory Treg** epitopes [1].
- **Results**: **Mutations gained after nivolumab therapy are less immunogenic and more tolerogenic** compared to mutations found prior to therapy.
- **Response to therapy is associated with Ancer results.**
- **Summary**: Our Ancer analysis suggests that nivolumab therapy affects the immunogenicity and tolerance profiles of newly generated mutations in a manner that is **consistent with the concepts of immunoediting and immune camouflaging**.

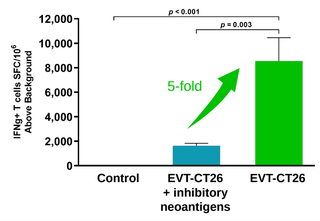
Background: Ancer® – The Answer to Cancer



Foreign vs self epitopes: *JanusMatrix* identifies **inhibitory (self-like) epitopes** *in silico*. Validation studies with pathogens [2, 3, 4] and cancer mutanomes [5].

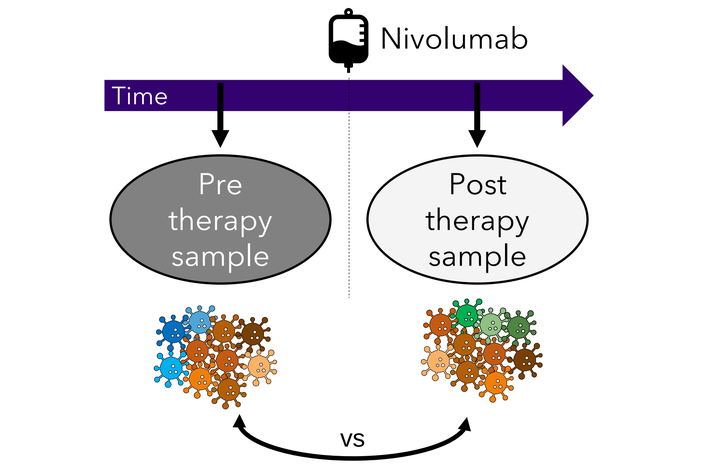


*CT26 self-like neoepitopes (**identified in silico with JanusMatrix**) suppress IFNg production to EVT-CT26 vaccines [5]*



Approach

- Forty-one pairs of mutanomes collected before (*Pre*) and while on (*Post*) nivolumab therapy were retrieved from [6].
- Pre and Post mutanomes were compared to identify deleted, maintained, and newly acquired mutations.
- Immunogenic and tolerance potentials were calculated for all mutations with Ancer.

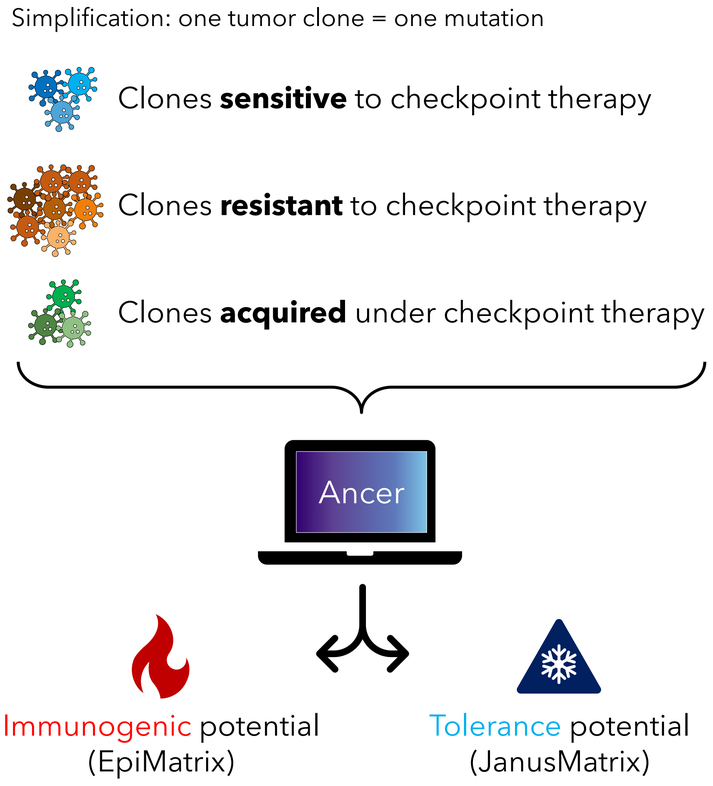


Simplification: one tumor clone = one mutation

Clones **sensitive** to checkpoint therapy

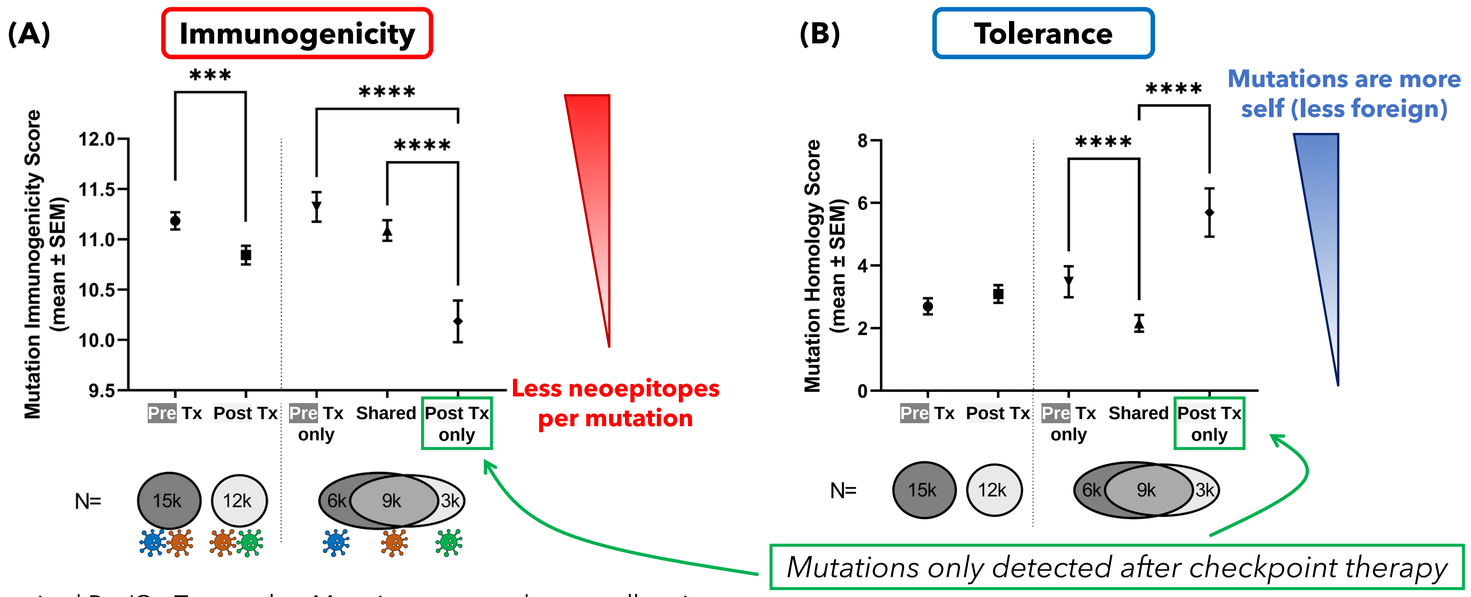
Clones **resistant** to checkpoint therapy

Clones **acquired** under checkpoint therapy



Results – Post-therapy mutations are less immunogenic and more tolerogenic

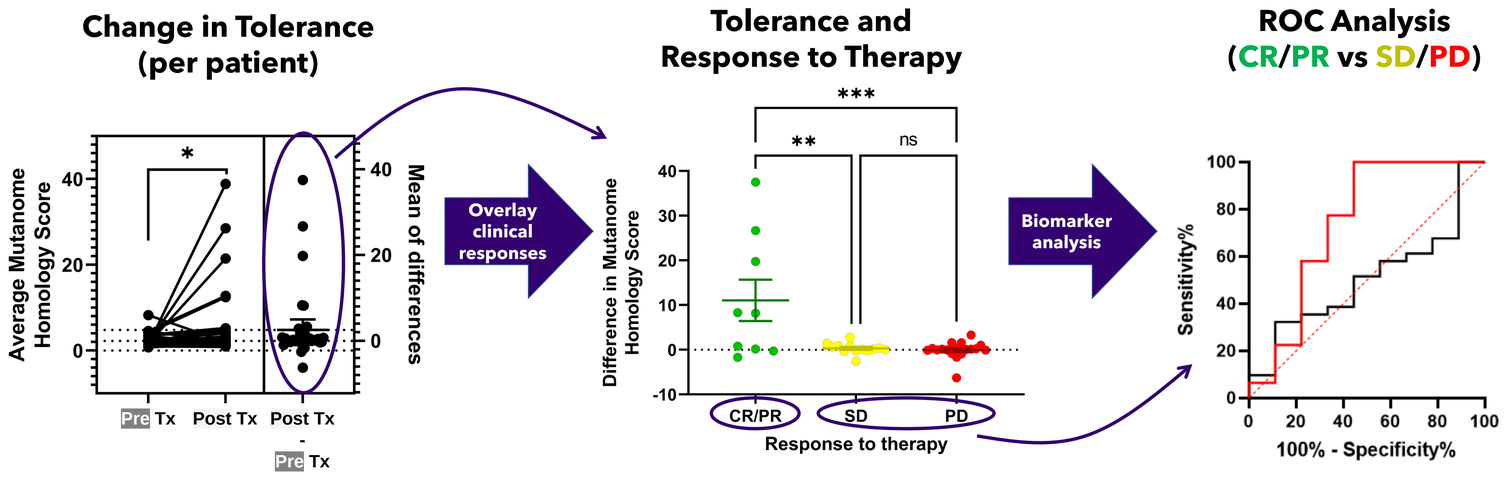
- Mutations gained after nivolumab therapy are less immunogenic (A) and more tolerogenic (B).
- Tumors respond to immunotherapy by reducing their immunogenicity and by avoiding the immune system.



N = 41 paired Pre/On Tx samples. Mutations grouped across all patients.
Pre Tx vs Post Tx: Mann-Whitney test. Pre Tx only vs Shared vs Post Tx only: ANOVA (Krustal-Wallis, multiple comparisons)

Results – Change in tumor tolerance is associated with response to therapy

- Tumors increase their tolerance potential (JanusMatrix homology scores) after nivolumab therapy.
- Change in tolerance is associated with response to therapy.



Change in JanusMatrix (homology) scores correlates with responses to therapy

	AUC	p-value
Baseline TMB	0.50	0.96
JanusMatrix	**0.74**	**0.03**

N = 41 paired Pre/On Tx samples. Samples paired by patient.
Pre Tx vs Post Tx: paired t test. CR/PR vs SD vs PD: ANOVA (multiple comparisons)
CR: Complete Response, PR: Partial Response, SD: Stable Disease, PD: Progressive Disease

Summary and Conclusions

- This study demonstrates **the utility of immunogenicity screening tools in the Ancer** platform for streamlined designs of personalized cancer vaccines.
- Our Ancer analysis suggests tumors reduce their immunogenicity (less neoepitopes) and increase their tolerance potentials (mutations more likely to be tolerated) in response to nivolumab therapy. **Mutations acquired after immunotherapy are more "stealth"** than mutations found prior to therapy.
- These results highlight that **identifying relevant mutations for precision immunotherapy (e.g. personalized vaccines) will become more difficult** once patients are treated with a checkpoint inhibitor. Specialized tools, such as JanusMatrix are needed to correctly and quickly identify immunogenic mutations.
- Ancer can be employed to **identify novel biomarkers** associated with clinical responses. Ancer **identified a tolerance signature** specific to patients who respond to nivolumab, suggesting Ancer can be used to triage patients for immunotherapy clinical trials.

References

1. Richard G.. et al., Expert Rev Vaccines. 2022 Feb;21(2):173-184
2. Liu R. et al., Hum Vaccin Immunother. 2015 11:9, 2241-2252
3. Wada Y. et al., Sci Rep. 2017 Apr 28;7(1):1283
4. Jang H. et al., Hum Vaccin Immunother 2020 Sep 1;16(9):2042-2050
5. Richard G. et al., Proceedings: AACR Annual Meeting 2019; March 29-April 3, 2019; Atlanta, GA
6. Riaz N. et al., Cell 2017; 171, 934–949