Enhancing TCR^{tg} T cell therapy efficacy in solid tumors using mRNA-based vaccination

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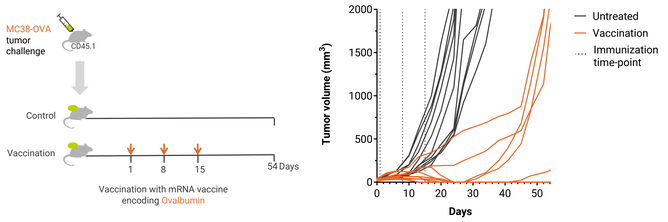
BACKGROUND AND AIMS

- In recent years, adoptive T cell therapy (ACT) has demonstrated remarkable efficacy against B-cell malignancies.
  - However, clinical benefit in solid tumors remains limited and short-lasting.
  - ACT faces several challenges in solid tumors such as inadequate tumor penetration and limited functionality and durability of the T-cell response.
- To address the limitations, we explored mRNA vaccination as a strategy to enhance the efficacy of TCR transgenic (TCR^{tg}) T cell ACT in the MC38-OVA mouse model.
  - We hypothesized that an OVA-mRNA vaccine could activate both transferred and endogenous T cells and thus enhance antitumor efficacy in comparison to ACT alone.

RESULTS

1. mRNA VACCINATION DELAYS TUMOR GROWTH

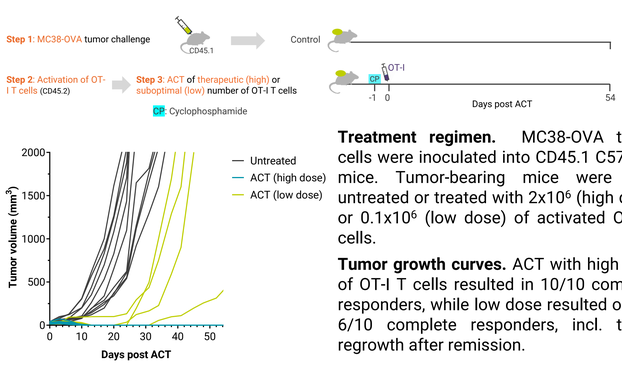
- mRNA vaccination against the model tumor antigen OVA temporarily delays MC38-OVA tumor growth, thus allowing assessment of potential synergies with ACT.



Treatment scheme. MC38-OVA expressing tumor cells were inoculated into CD45.1 C57BL/6 mice. Tumor-bearing mice were kept untreated or vaccinated with a mRNA-based vaccine encoding Ovalbumin.

2. IDENTIFICATION OF SUBOPTIMAL T CELL DOSE FOR ACT

- Adoptive transfer of a low dose of activated ova-specific OT-I TCR^{tg} T cells has limited anti-tumoral efficacy in MC38-OVA model.

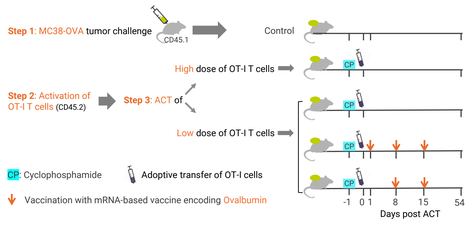


Treatment regimen. MC38-OVA tumor cells were inoculated into CD45.1 C57BL/6 mice. Tumor-bearing mice were kept untreated or treated with 2x10^6 (high dose) or 0.1x10^6 (low dose) of activated OT-I T cells.

Tumor growth curves. ACT with high dose of OT-I T cells resulted in 10/10 complete responders, while low dose resulted only in 6/10 complete responders, incl. tumor regrowth after remission.

3. COMBINATION OF ACT AND EARLY VACCINATION INDUCES AN EFFICIENT AND LONG-LASTING ANTI-TUMORAL RESPONSE

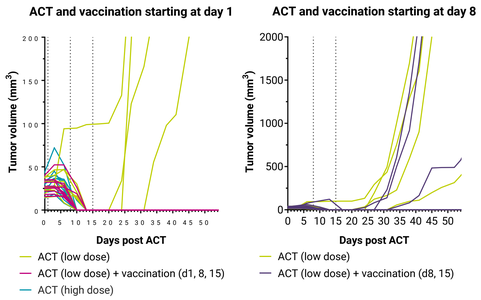
- Transfer of a low dose of OT-I T cells combined with early vaccination (one day after ACT) resulted in complete tumor regression with long-lasting efficacy.
- In contrast, transfer of a low dose of OT-I T cells combined with late vaccination (eight day after ACT) resulted in tumor regression in most but not all mice.



Treatment regimen.

MC38-OVA tumor cells were inoculated into CD45.1 C57BL/6 mice.

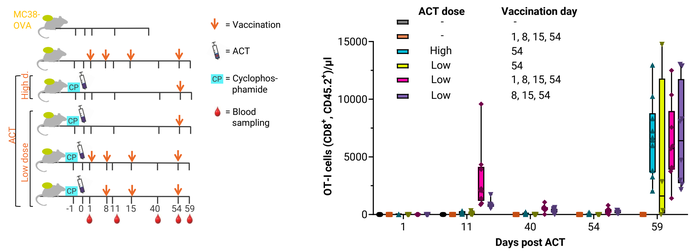
Tumor-bearing mice were kept untreated or treated with 0.1x10^6 (low dose) or 2x10^6 cells (high dose) activated OT-I T cells (CD45.2$^+$).



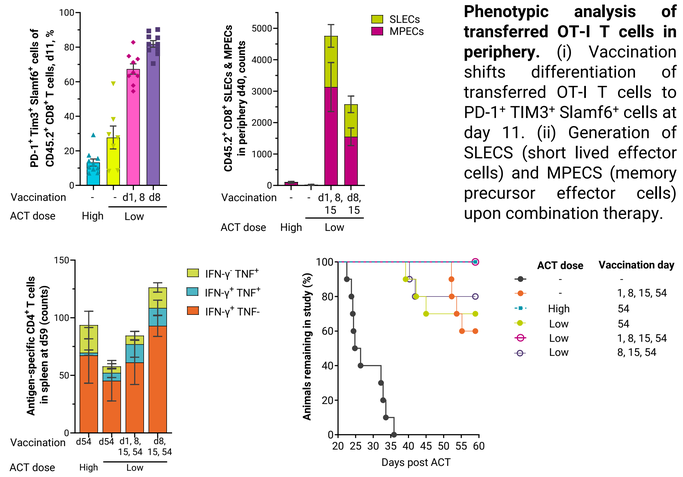
Tumor growth curves. (i) Low-dose ACT with vaccination starting one day post ACT achieved tumor regression in all mice, while low-dose ACT alone resulted in 6/10 complete responders. High-dose ACT achieved also tumor regression in all mice. (ii) Vaccination starting eight days post ACT eradicated tumors in 7/10 mice.

4. VACCINATION EXPANDS TRANSFERRED OT-I T CELLS IN BLOOD

- Vaccination increases number of transferred OT-I T cells in the periphery.
- Vaccination 54 days *post* ACT expands transferred T cells both in mice that have already been vaccinated multiple times after ACT and mice that have not been vaccinated after ACT.
- Early vaccination + low ACT dose yields 100% survival in the MC38-OVA model.



Transferred OT-I T cells in the periphery and mouse survival. Low amount of transferred OT-I T cells could be boosted by vaccinations (day 11, pink and violet bar). 25 days *post* last vaccination, the OT-I T cells were not anymore observed in the circulation (day 40). Vaccination at day 54 again expanded the transferred OT-I T cells (day 59, pink and violet bar). One vaccination 54 days *post* ACT could expand peripheral OT-I T cells in mice (day 59, blue and yellow bars).

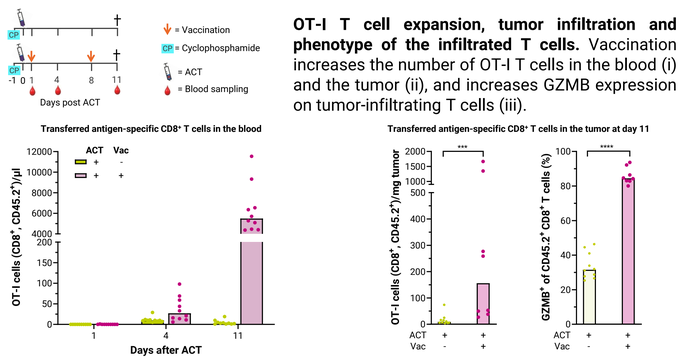


Phenotypic analysis of transferred OT-I T cells in periphery. (i) Vaccination shifts differentiation of transferred OT-I T cells to PD-1$^+$ TIM3$^+$ Slamf6$^+$ cells at day 11. (ii) Generation of SLECS (short lived effector cells) and MPECS (memory precursor effector cells) upon combination therapy.

Analysis of endogenous antigen-specific CD4$^+$ T cells in the spleen and therapy efficacy. Polyfunctional antigen-specific CD4$^+$ T cells (teal) are mainly expanded after multiple vaccinations (iii). Early OVA-mRNA vaccination (starting day 1) rescued the efficacy of the low OT-I dose (iv).

5. VACCINATION INCREASES INFILTRATION AND CYTOTOXICITY OF THE TRANSFERRED T CELLS

- Vaccine-mediated increase in number of OT-I T cells in the periphery is accompanied by increase of OT-I T cells in the tumor.
- Vaccination promotes a cytotoxic phenotype in the tumor infiltrating OT-I T cells.



OT-I T cell expansion, tumor infiltration and phenotype of the infiltrated T cells. Vaccination increases the number of OT-I T cells in the blood (i) and the tumor (ii), and increases GZMB expression on tumor-infiltrating T cells (iii).

CONCLUSION

- Our preclinical studies investigated the effects of mRNA vaccination on transferred TCR^{tg} and endogenous T cells, as well the impact of vaccination on tumor growth, mouse survival and T cell infiltration into the tumors.
- The obtained data demonstrate a significant improvement in adoptive TCR^{tg} T cell therapy efficacy, when combined with an mRNA vaccine.

KEY TAKEAWAYS

- ACT with CAR T cells has revolutionized the treatment of B cell malignancies, and ACT using TCR^{tg} T cells has shown promise in several cancer types.
- ACT of solid cancers suffers from insufficient infiltration, effector functions and persistence of the T cells.

- This preclinical work utilized the MC38-OVA mouse tumor model to test whether the efficacy of TCR^{tg} T cell ACT could be increased by mRNA vaccination.

MC38-OVA
+ OT-I ACT
+ OVA-mRNA based vaccine

- Tumor growth was suppressed, and mouse survival increased by the addition of an mRNA vaccine into the ACT regimen.
- Vaccine promoted expansion, infiltration and effector phenotype of the transferred T cells.
- Vaccination also increased the number of endogenous, antigen specific polyfunctional CD4+ T cells.

- Combining mRNA vaccination with ACT **increased therapy efficacy** in comparison to monotherapies and established a long lasting immunosurveillance.
- The combinatorial approach paves way for **improved ACT outcomes** against solid tumors.

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References

1. Kirtane *et al.*, J Immunother Cancer 2021 Jul;9(7):e002723. doi: 10.1136/jitc-2021-002723.

Acknowledgements

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