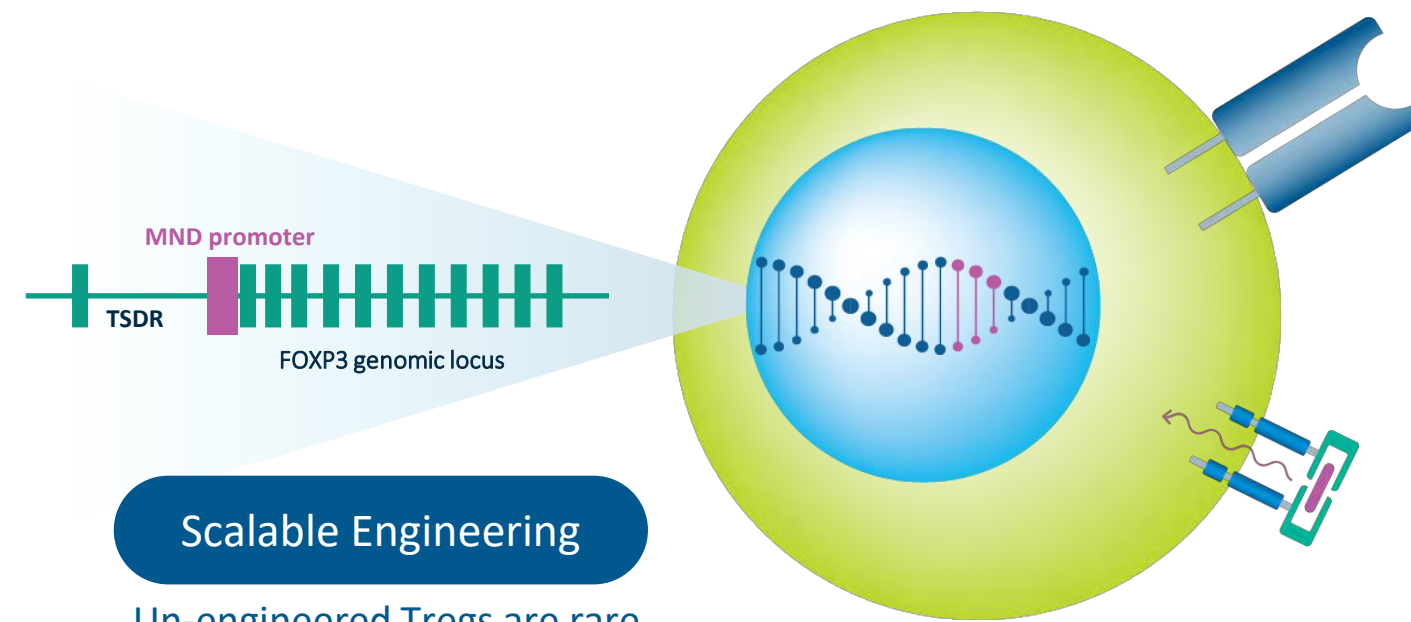


Payam Zarin, Gene I Uenishi, Martina Sassone-Corsi, Tingxi Guo, Victoria DeVault-Nelson, Maegan Hoover, Jennifer Yam, Nathan Zammit, Abigail Doherty, Priya Saikumar-Lakshmi, Yash Agarwal, Madison Milaszewski, Nicole Reed, Jennifer Mellen, David Tucker, Ashley Landuyt, Carlos E Frias, Alaina Burgess, Sophia Hernandez, Alberto Del Rio-Espinola, Dalia Gaddis, Chris Moore, Xiaoming Hu, Jun Chen, Marko Repic, Mark Bach, Tiffany F Chen, Thomas Wickham
 GentiBio, Inc., Cambridge, MA, USA

Premise

Engineered Tregs address key shortcomings of sorted Treg approach



Scalable Engineering

Un-engineered Tregs are rare and heterogenous

- Tregs engineered from more abundant bulk T-cells are scalable and achieve better phenotypes by stabilizing Treg differentiation factors (e.g. FOXP3) from either autologous or allogeneic sources.

Tissue Targeting

Additional transgene enables tissue specificity

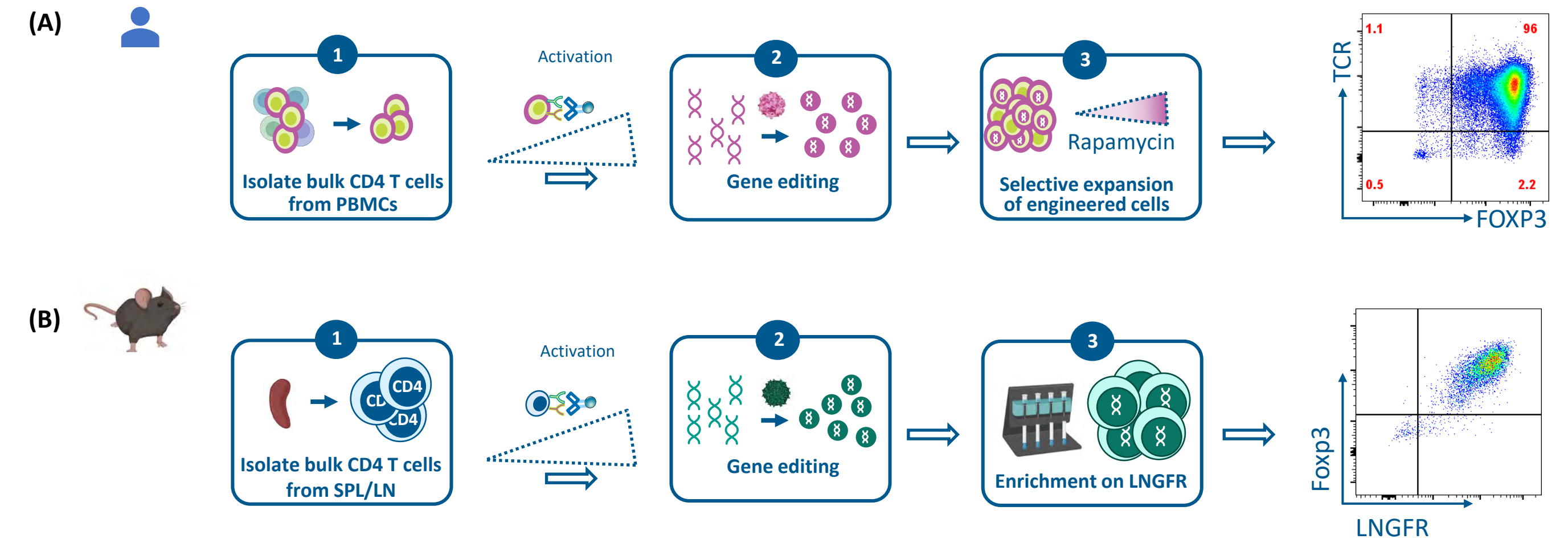
- Modular TCRs or CARs for optimal efficacy and safety
- Antigen specificity confers tissue localization, engages bystander suppression and drives infectious tolerance
- Genti approach to TCR & CAR screening and qualification identifies TCRs & CARs that are optimal for pairing with EngTregs

IL2 Signaling

Tregs require IL2 but do not produce it

- Engineered Tregs incorporate Treg-selective and titratable IL2 signaling through a chemically-induced signaling complex (CISC)
- CISC enhances manufacturing and transforms Tregs into potent, long-lived drugs

Engineered Treg generation

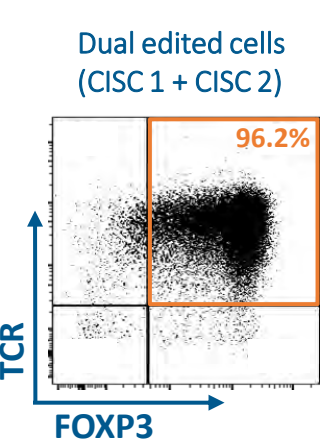
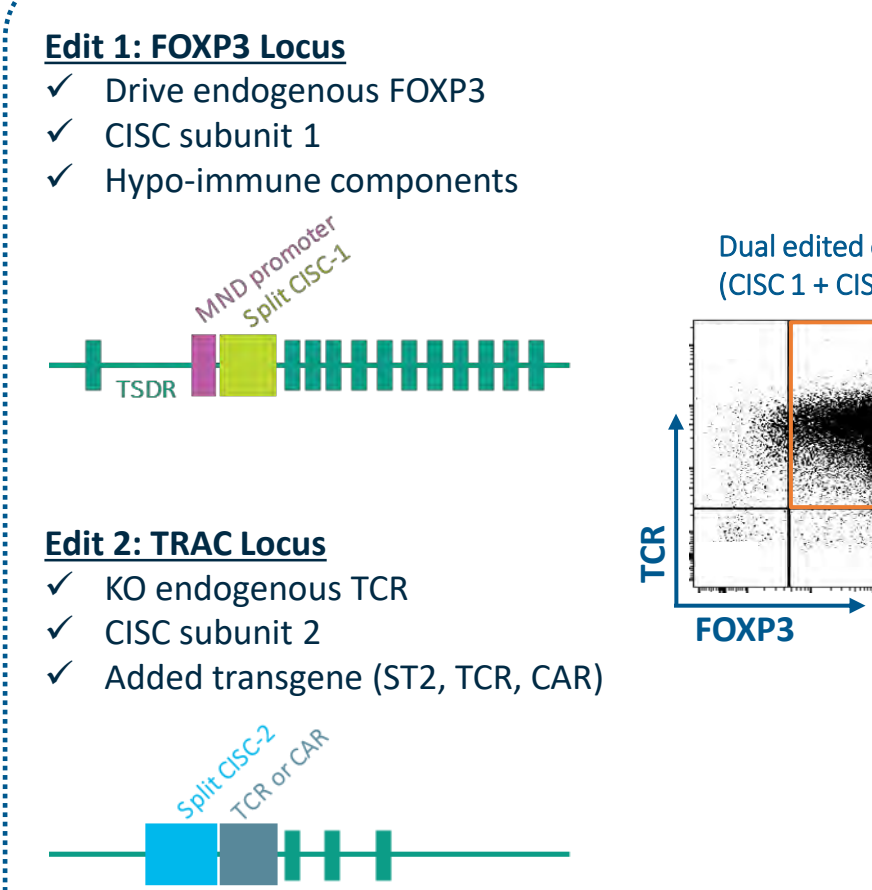


Limitations of expanding sorted natural regulatory T cell. Natural Tregs are a rare subset within the PBMCs of most individuals. While the expansion of these rare cells in culture following cell sorting has been a common approach over the past decade, several well documented, key inherent limitations involving stability of natural Tregs have come to light (Zhou X., *Nat. Immunol.*, 2009; Junius S., *Sci. Immunol.*, 2020).

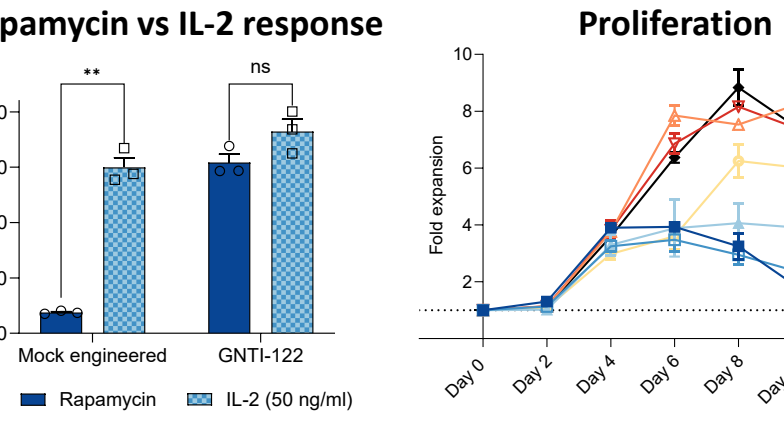
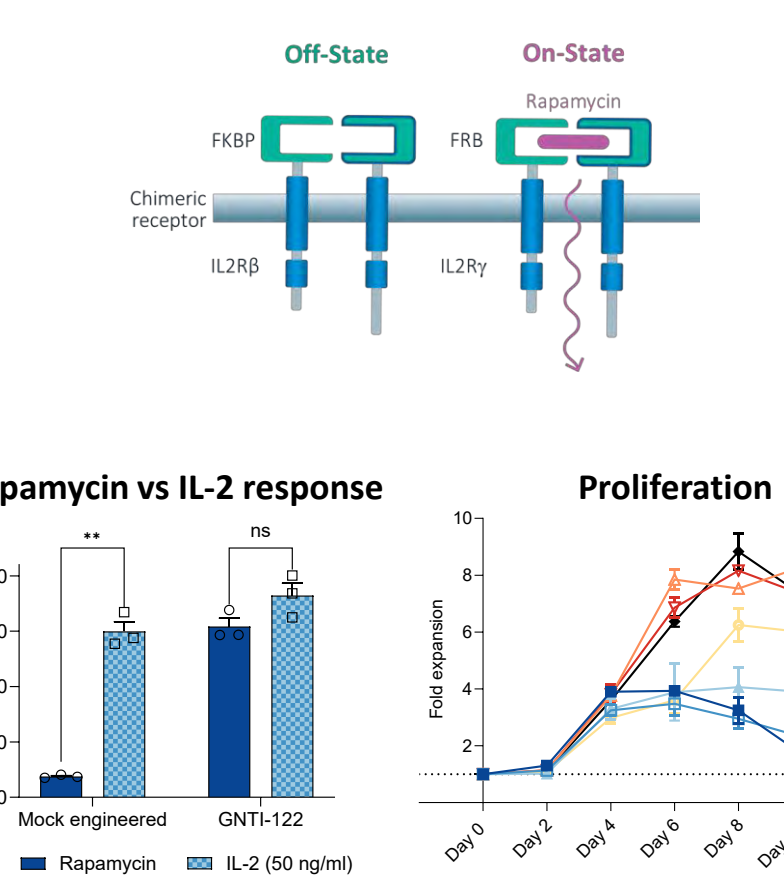
Engineered Treg production processes. A) Human or (B) murine CD4+ T cells are isolated (Panel 1) and activated via α -CD3/ α -CD28 microbeads. Cells are genetically modified using CRISPR-Cas9 mediated knock-in of transgenes delivered by AAV vectors (Panel 2). For human EngTregs, CISC receptor expression enables selective expansion of edited cells, while mouse surrogate EngTregs rely on enrichment by the extracellular LNGFR tag (Panel 3).

Highly scalable manufacturing results in stable Tregs without the need for complex sorting

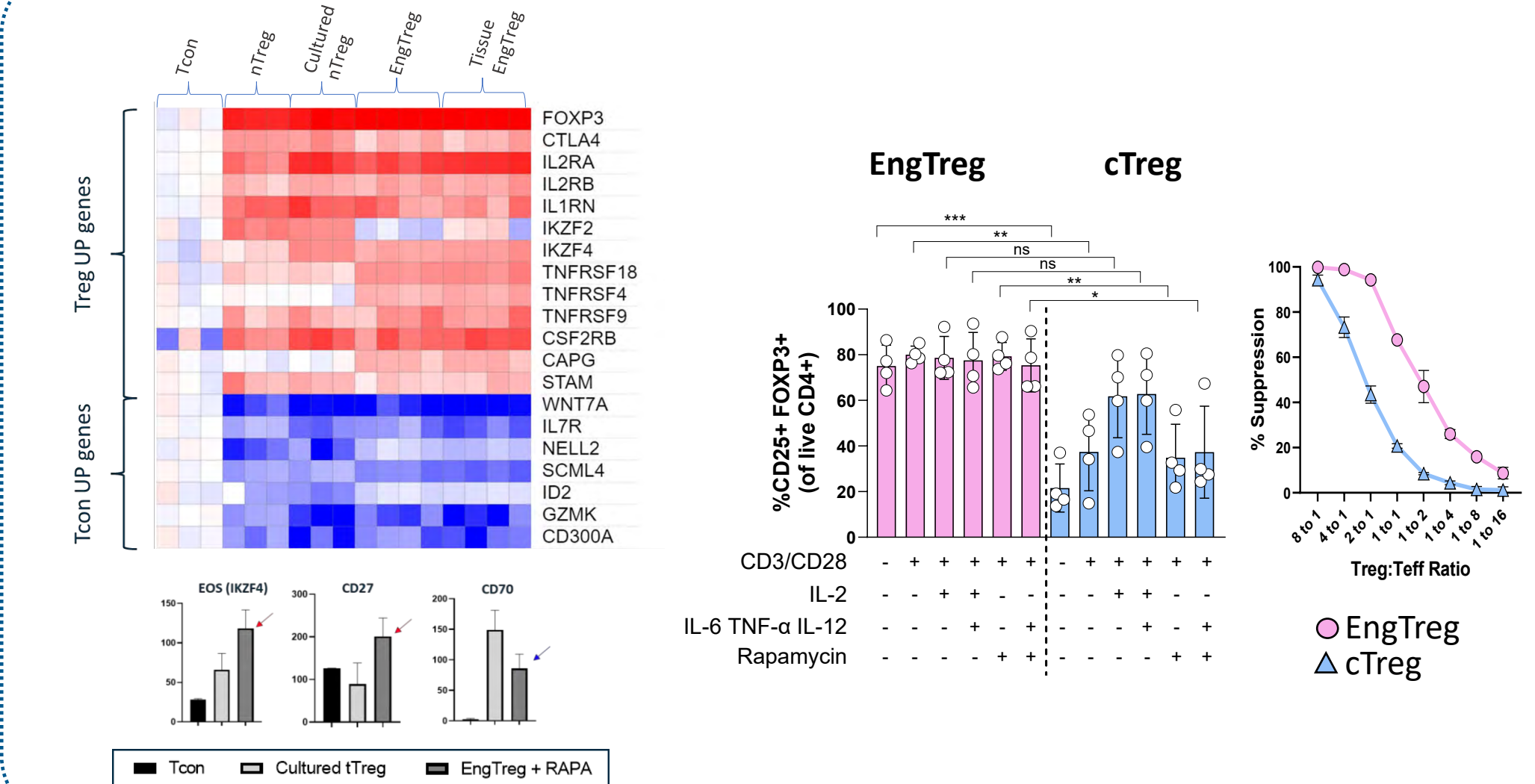
Dual edit strategy yields pure EngTreg product



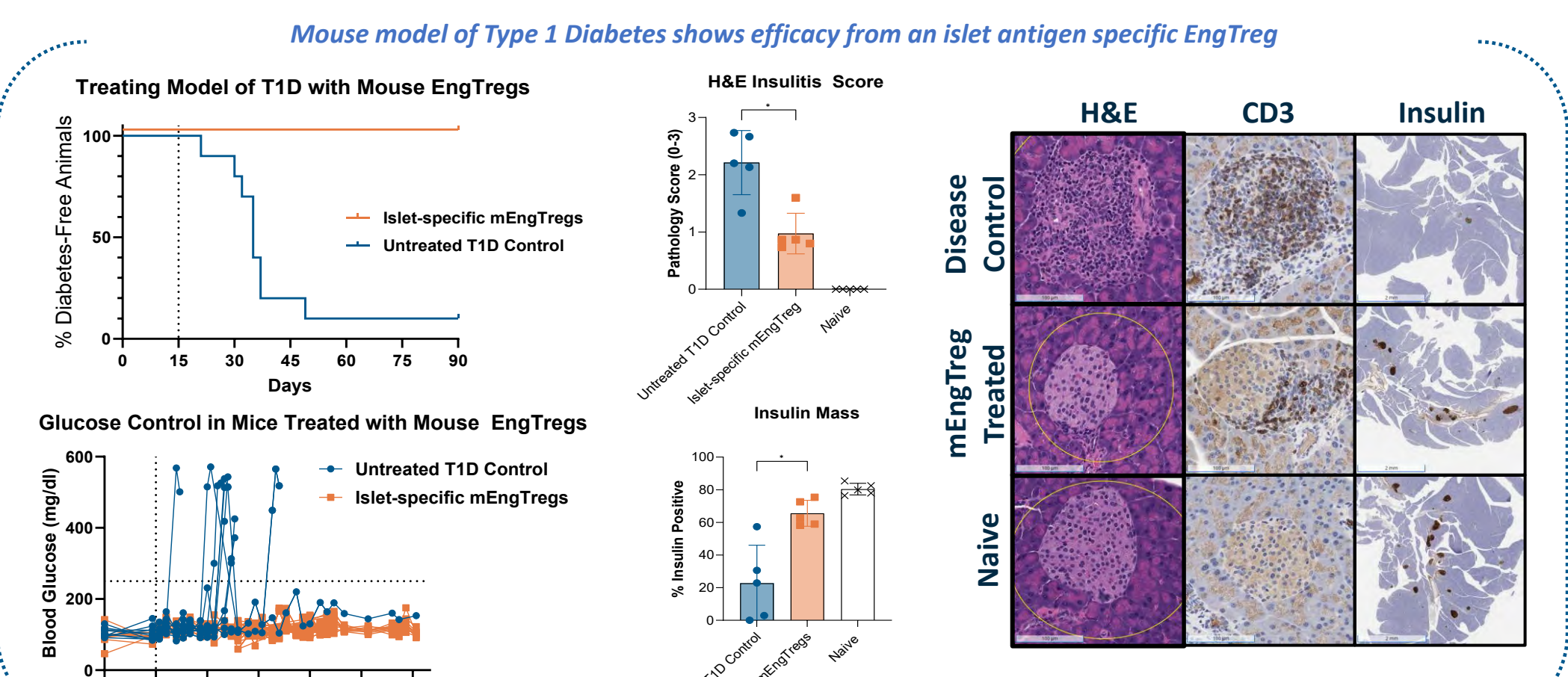
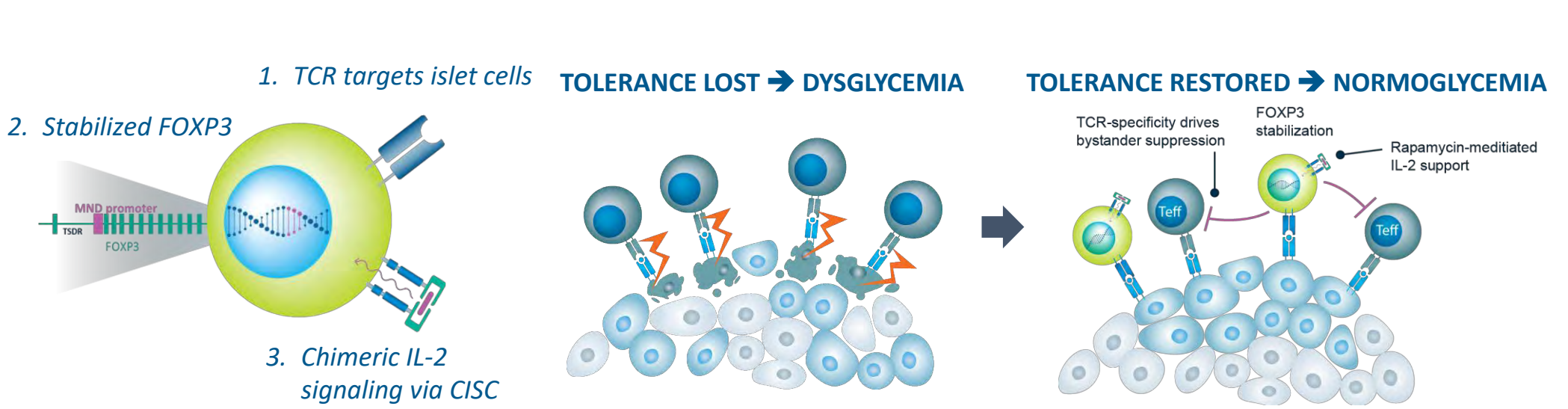
CISC imparts a selective, tunable IL-2 like signal



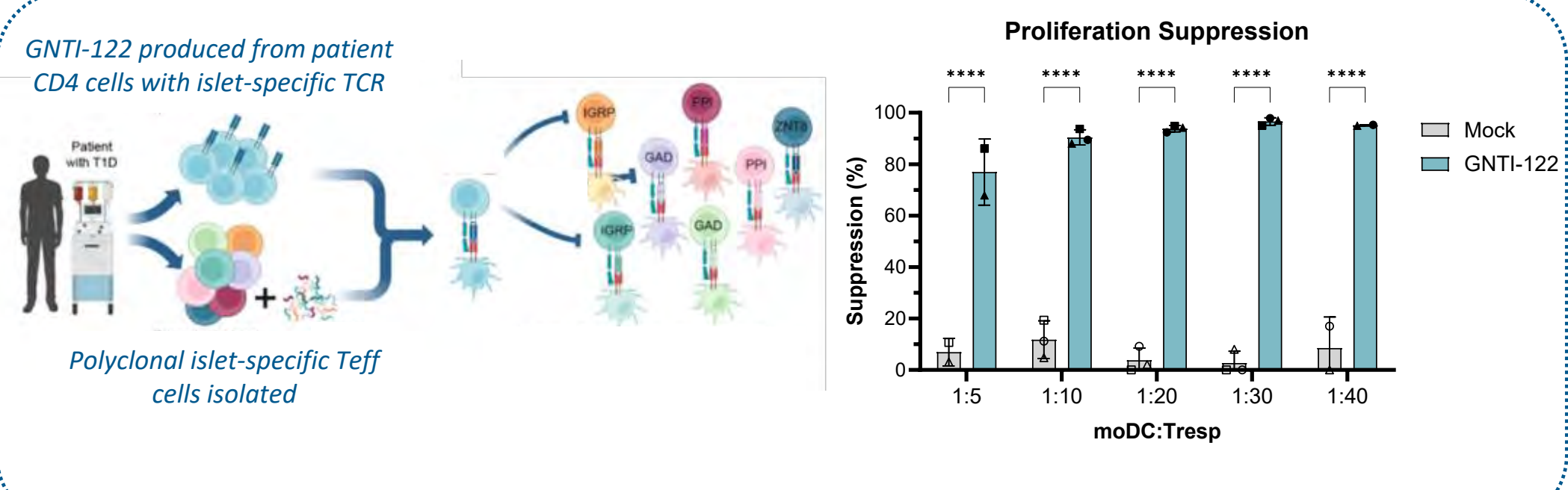
Engineered Tregs maintain higher stability and suppressive capacity compared to cultured nTregs (cTregs)



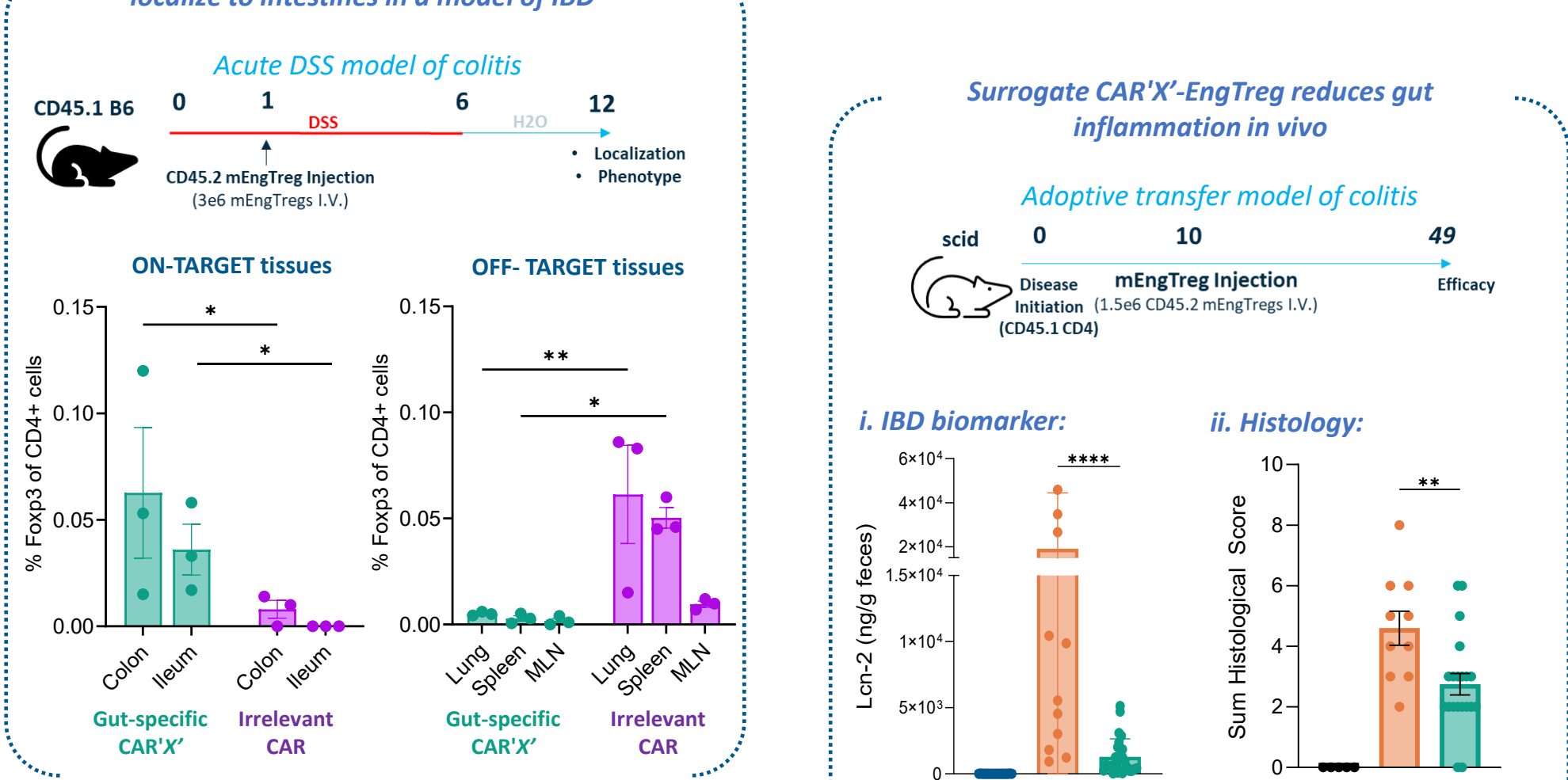
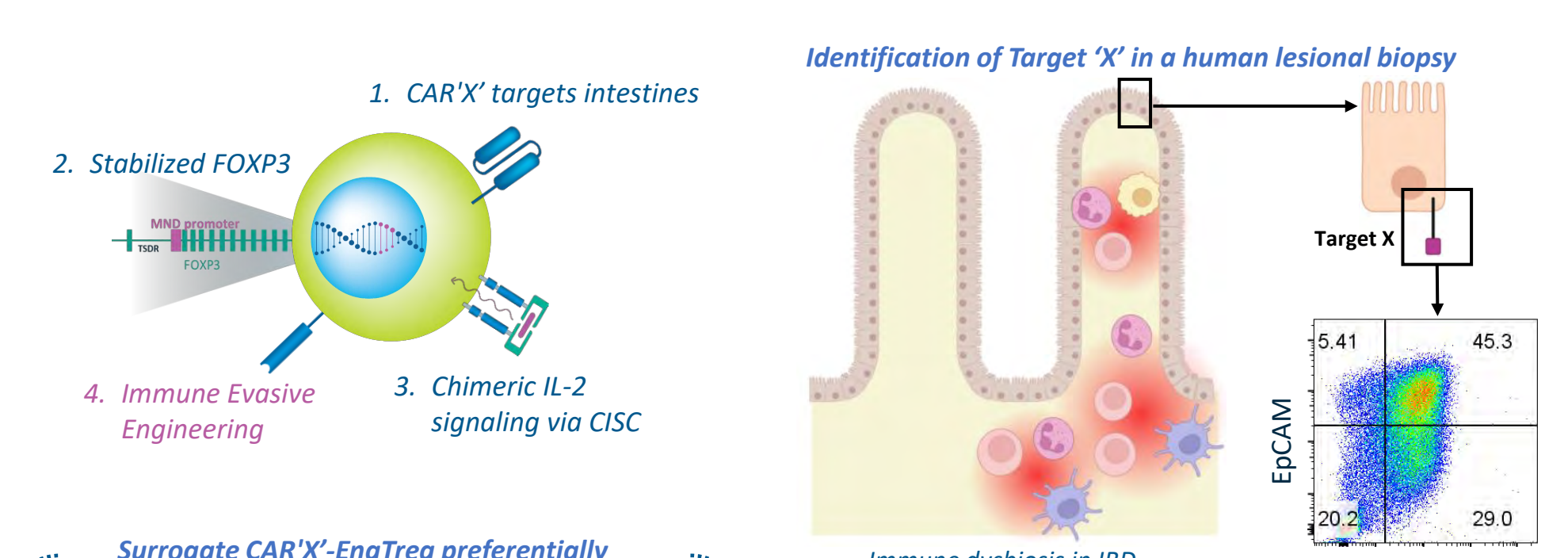
GNTI-122 suppresses pathogenic islet-specific Tregs that cause T1D



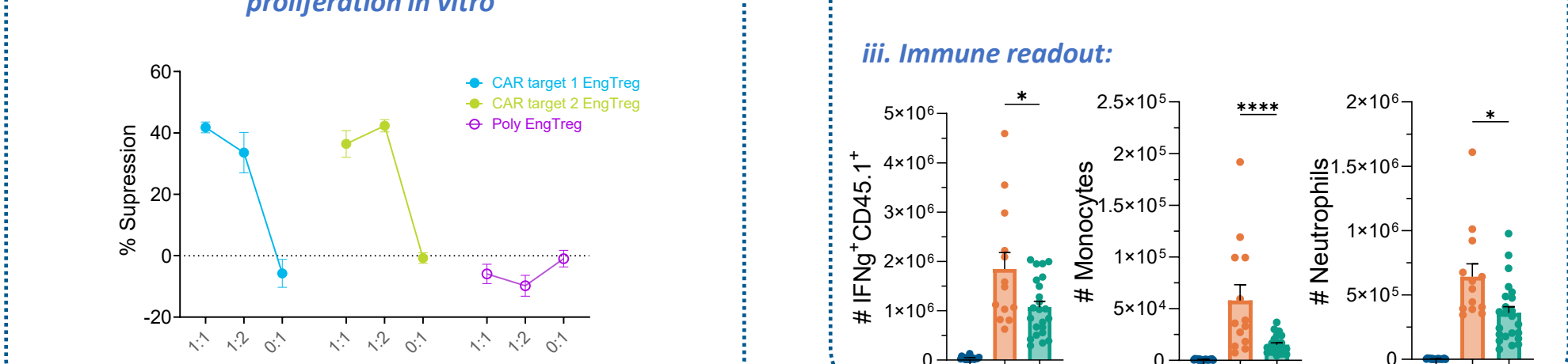
GNTI-122-mediated suppression of polyclonal islet Treg cells from same patient



GNTI-932 Hypo-immune (off-the-shelf) allogeneic EngTreg for IBD

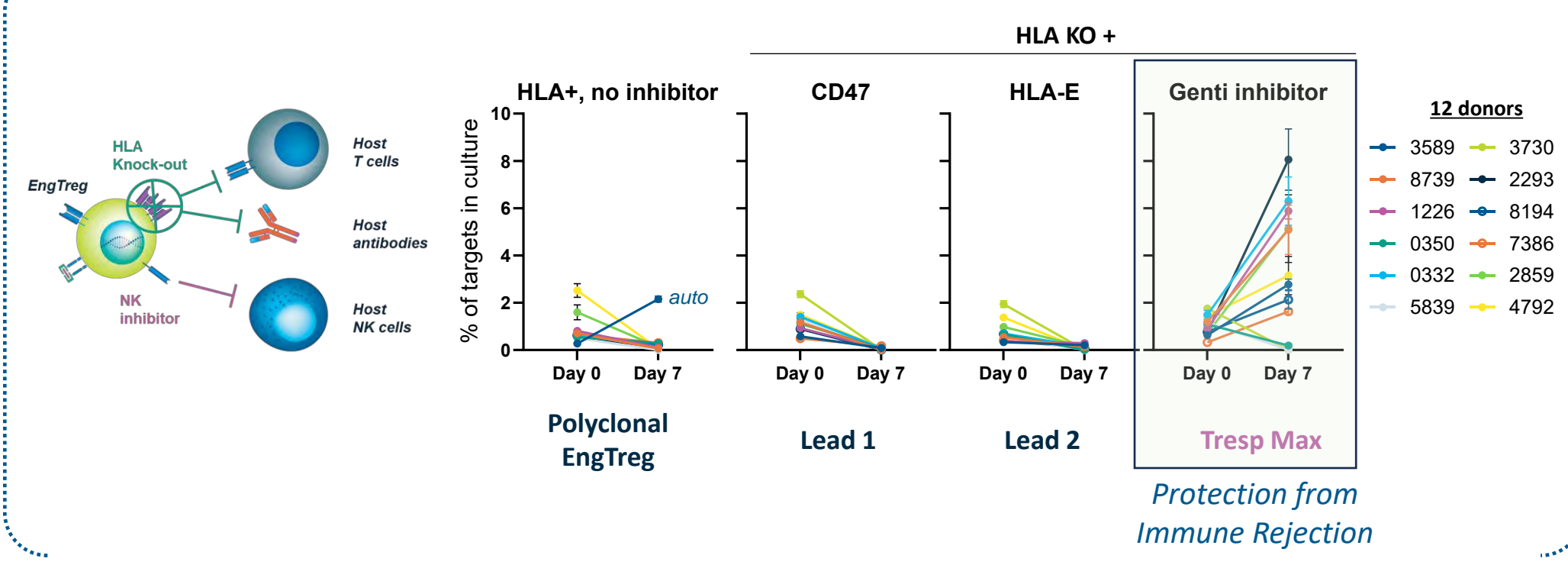


CAR 'X'-dependent suppression of T-eff proliferation in vitro

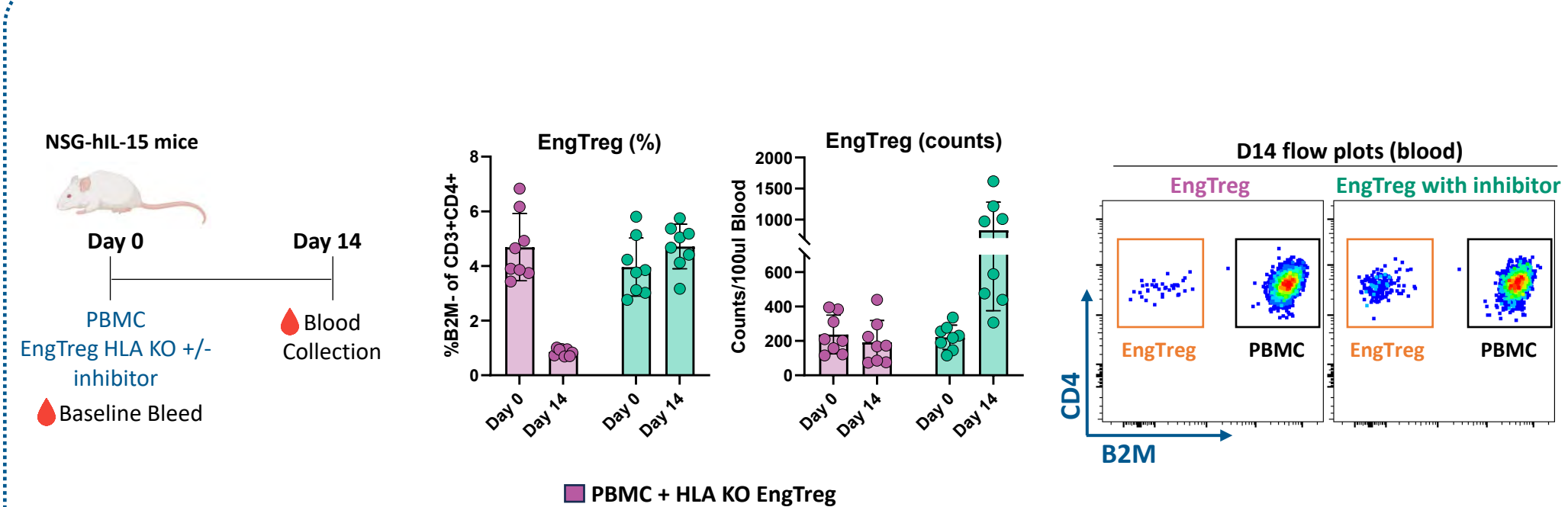


Best in class proprietary Hypo-immune Engineering enables allogeneic persistence

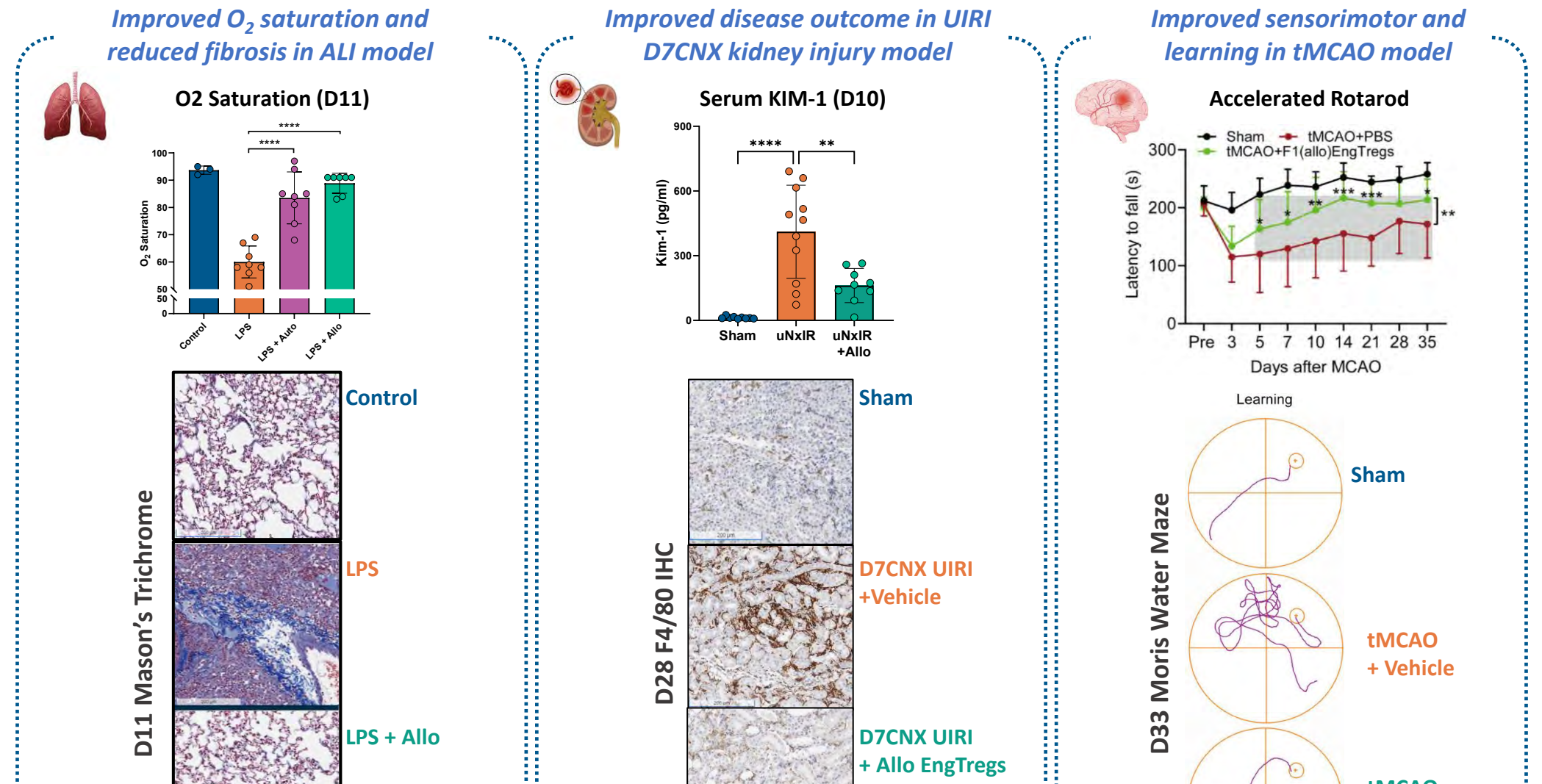
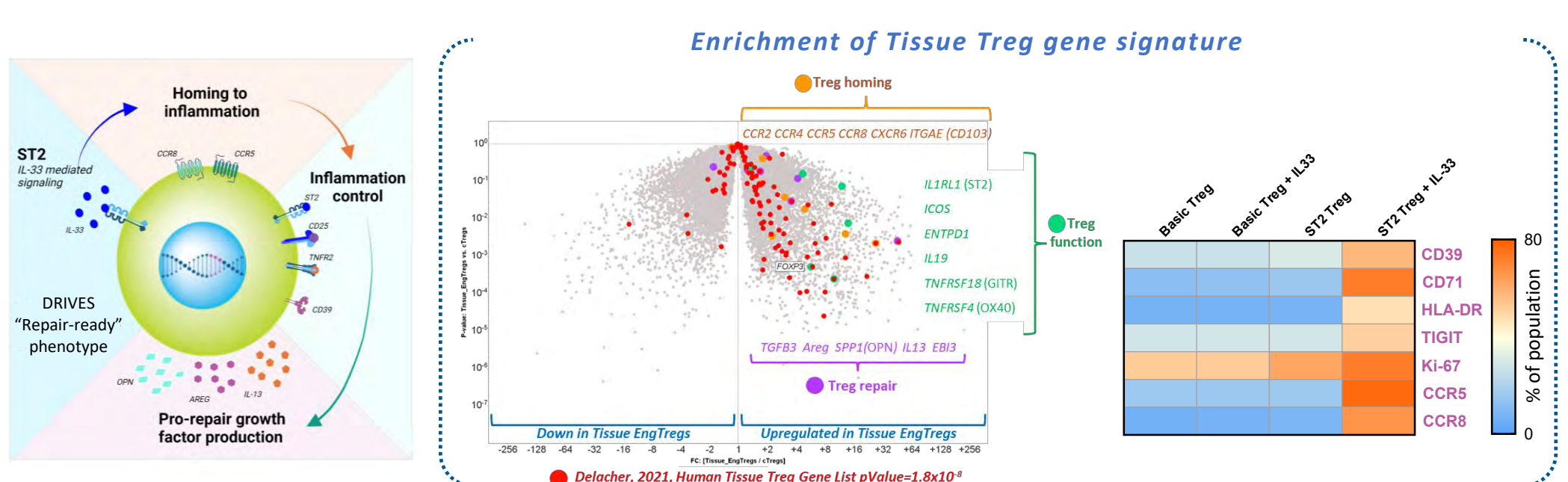
In vitro support of immune evasion technology



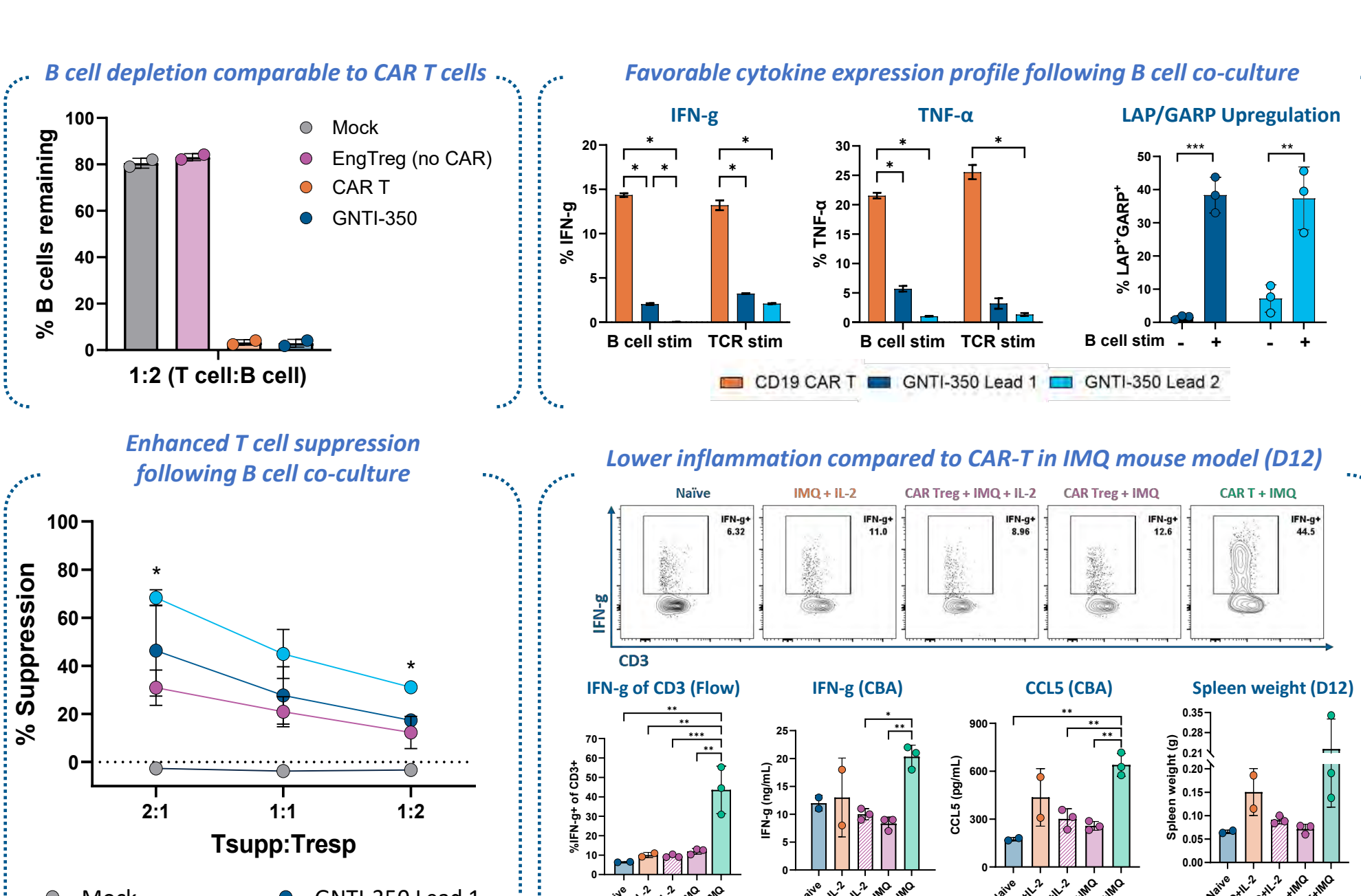
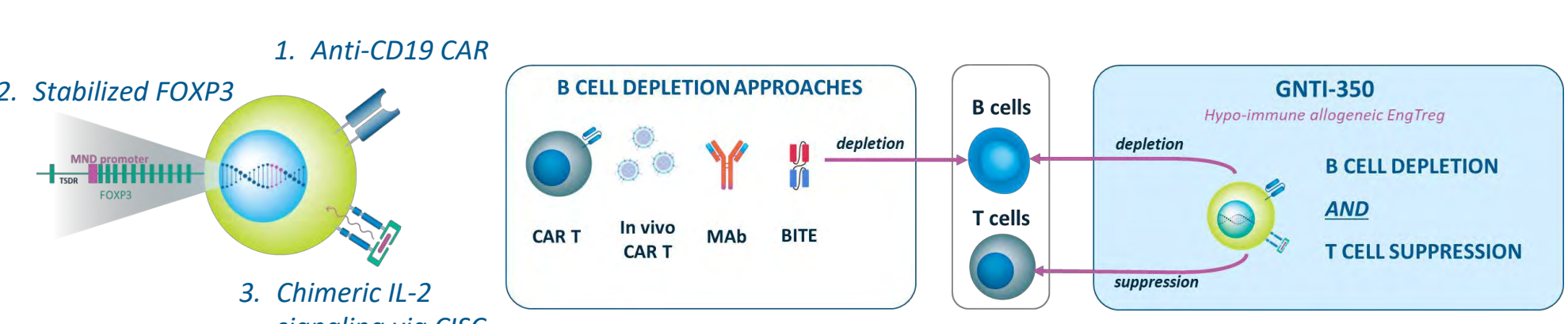
In vivo proof of concept showing IEE driven protection of EngTregs in the NSG-HL15 model with PBMC transfer



GNTI-823 curtails acute inflammation and supports tissue repair



GNTI-350 shows improved immune reset and safety versus CAR T



CONCLUSIONS

- GentiBio's Engineered Treg platform overcomes scaling and stability limitations of Treg therapeutics by starting with more abundant T cell sources and enriching FOXP3+ edited cells with an engineered IL-2 signaling receptor.
- Engineered Tregs express higher levels of Core Treg genes such as FOXP3, IL2RA and CTLA4 as well as FOXP3 Synergy genes, such as IRF4 and EOS, compared to cTregs at both the bulk and single cell RNA transcript levels.
- GentiBio's Hypo-immune Engineering enables allogeneic Treg products with long persistence, low COGs, and the potential for repeat dosing.
- GNTI-122 suppresses pathogenic T1D-driving islet specific Tregs; murine surrogate EngTregs preserve islets and prevent diabetes onset.
- GNTI-823 Engineered Tissue Tregs express high levels of tolerogenic and repair mediators; murine surrogate EngTregs are efficacious in models of acute lung injury, kidney injury, and ischemic stroke.
- GNTI-932 selectively binds to a gut epithelial target, showing enhanced homing and efficacy in the context of IBD pathology.
- GNTI-350 depletes B cells, induces comprehensive immune reset, and displays a better safety profile compared to CD19 CAR T cells.

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*, **, ***, **** = p-value < 0.01, 0.005, 0.001 and 0.0001 respectively.

We make Tregs. Better.

