POWERED BY genOway's PRECLINICAL MODELS

Featuring the genO-hCCR8/hCCL1 Scan or click

Featuring the genO-hFcγR Scan or click

Featuring the genO-BRGSF-HIS Scan or click

genOway ULTIMATE PREDICTABILITY

genO-hCCR8/hCCL1/hFcγR preclinical mouse model for efficacy assessment of CCR8-targeting agents

Fabiane SÔNEGO, Angela PAPPALARDO, Gaelle MARTIN, Julie GORRY, Yacine CHERIFI, Patricia ISNARD-PETIT, Kader THIAM
genOway, Lyon, 69007, France

SITC 2026 Abstract #401

Background: CCR8 is an emerging therapeutic target in oncology due to its role in Treg-mediated immunosuppression. The development of physiologically relevant preclinical models to evaluate CCR8-targeting therapies requires preservation of functional interactions between the receptor and its ligands. CCR8 has two known ligands, CCL1 and CCL8. Previous studies have shown that mouse CCL8 can induce calcium flux in human CCR8-transfected murine cells [1], suggesting that humanization of CCL8 may not be required. In contrast, murine CCL1 does not interact with human CCR8 in vitro, indicating that disruption of the CCR8/CCL1 axis may impair Treg suppressive function and thereby necessitate humanization of both CCR8 and CCL1.

In addition, depletion of CCR8$^+$ Tregs by therapeutic agents has been shown to depend on Fcγ receptor (FcγR) engagement [2]. However, significant interspecies differences in FcγR repertoire and expression pattern between mice and humans limit the translational relevance of mouse models expressing mouse FcγR. A previously described humanized FcγR mouse model expressing human FcγRI, FcγRIIA, FcγRIIB, FcγRIIIA and FcγRIIIB in a human-like distribution enables more accurate evaluation of Fc-mediated responses [3]. Building on this, we developed a novel humanized model expressing humanized CCR8, CCL1 and FcγRs (genO-hCCR8/hCCL1/hFcγR).

1. genO-hCCR8/hCCL1 HUMANIZED MOUSE MODEL

CCR8 and CCL1 were humanized by replacing the murine genes with their human counterparts under the control of endogenous mouse promoters. Physiological regulation of gene expression was preserved as no regulatory elements were altered during gene editing.

As presented during SITC 2025, genO-hCCR8/hCCL1 model has shown:
- No alteration of immune cell distribution compared to WT mice
- Detection of human CCL1 in the serum of genO-hCCR8/hCCL1 mice
- CCR8 expression on tumor-infiltrating Treg in MC38-engrafted mice
- CCR8-targeting depleting antibody treatment resulted in significant antitumor activity with reduced tumor volume and improved survival

2. genO-hFcγR HUMANIZED MOUSE MODEL

genO-hFcγR model was developed to express humanized:
- FcγRI
- FcγRIIAR131
- FcγRIIB
- FcγRIIIAF158
- FcγRIIIB

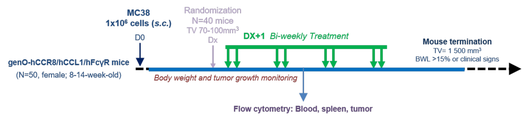
Expression of mouse FcγRI, FcγRIIB, FcγRIII and FcγRIV is abrogated.

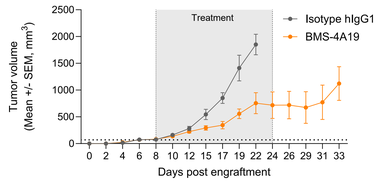
genO-hFcγR model has shown:
- FcγR expression overall follows a human-like expression pattern
- FcγR expression is regulated by cytokines as described in humans [3]
- ADCC is functional in NK cells
- Fc engagement correlates with effector functions demonstrated in auto-immunity, inflammatory diseases, immuno-oncology and infectious diseases [4]
- Capacity to mount robust antigen-specific T cell responses
- Development of Malaria and similar infectivity to C57BL/6N WT mice

✓ **genO-hFcγR model was intercrossed with the genO-hCCR8/hCCL1 model to generate the triple humanized genO-hCCR8/hCCL1/hFcγR strain**

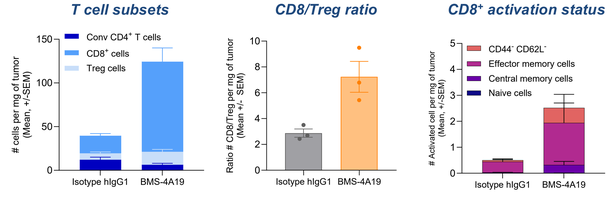
3. genO-hCCR8/hCCL1/hFcγR HUMANIZED MOUSE MODEL FOR EFFICACY ASSESSMENT OF CCR8-TARGETING THERAPIES

A. ANTI-CCR8 ANTIBODY INHIBITS TUMOR GROWTH IN genO-hCCR8/hCCL1/hFcγR model





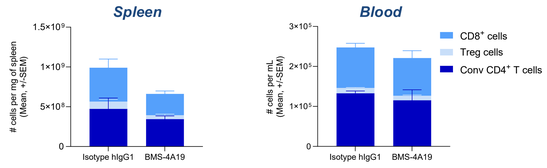
B. INTRATUMORAL TREG AND CD8$^+$ T-CELL DISTRIBUTION AND PHENOTYPE



✓ **Under the tested conditions, BMS-4A19 induces recruitment of T-cells in the TME and an increase of CD8/Treg ratio**

✓ **Increased CD8/Treg ratio is associated with both effector and central-memory CD8$^+$ T-cell phenotypes**

C. T-CELL DISTRIBUTION IN SPLEEN AND BLOOD



✓ **T-cell subset distribution remains comparable in both spleen and blood**

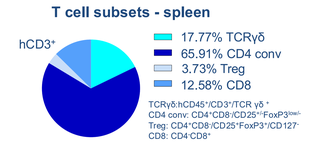
✓ **However, CCR8 expression pattern differs between human and mouse as human CCR8 is also expressed on peripheral Tregs**

✓ **Safety assessment requires a model with human-like expression of CCR8**

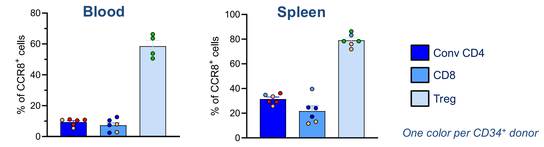
4. genO-BRGSF-HIS MODEL FOR SAFETY ASSESSMENT OF CCR8/CCL1-TARGETING THERAPIES

A. genO-BRGSF-HIS MODEL FEATURES

BRGSF (BALB/c *Rag2*$^{-/-}$, *IL2Rγ*$^{-/-}$, *SIRPa*^{NOD}, *Flt3*$^{-/-}$):
- Highly immunodeficient with reduced murine myeloid cells
- Normal lifespan (no anemia, no weight loss, normal fur texture & integrity, normal activity & posture)
- Upon CD34$^+$ cells injection:
  - Lymphoid and Myeloid compartment development

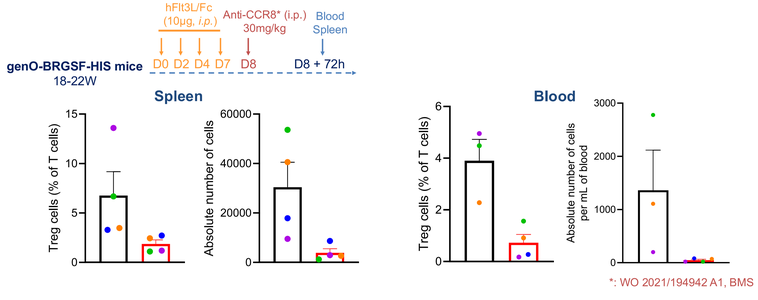


B. IN genO-BRGSF-HIS, CCR8 IS EXPRESSED BY NAÏVE TREG IN BLOOD AND SPLEEN AS IN HUMANS



One color per CD34$^+$ donor

C. ANTI-CCR8 DEPLETES TREG IN SPLEEN AND BLOOD OF NAÏVE genO-BRGSF-HIS



*: WO 2021/194942 A1, BMS

Conclusion: The genO-hCCR8/hCCL1 model represents a valuable platform for assessing the efficacy of CCR8-targeting therapies and investigating immunomodulatory mechanisms within the tumor microenvironment. The addition of humanized FcγRs in the genO-hCCR8/hCCL1/hFcγR model further enhances translational relevance by enabling evaluation of Fc-mediated effector functions. Nevertheless, this model has limitations to assess safety of CCR8-targeting compounds as the pattern of expression differs between human and mouse. The human-like expression pattern of CCR8 in naïve Treg from genO-BRGSF-HIS model makes it a more suitable model for safety and PD assessment.

Acknowledgement: Abilita Therapeutics

www.genoway.com

References:

[1] Islam SA, Chang DS, Colvin RA, et al. Mouse CCL8, a CCR8 agonist, promotes atopic dermatitis by recruiting IL-5$^+$ T(H)2 cells. Nat Immunol. 2011;12(2):167-177. doi:10.1038/ni.1984

[2] Campbell JR, McDonald BR, Mesko PB, et al. Fc-Optimized Anti-CCR8 Antibody Depletes Regulatory T Cells in Human Tumor Models. Cancer Res. 2021;81(11):2983-2994. doi:10.1158/0008-5472.CAN-20-3585

[3] Van Damme KFA, Sichien D, Van der Borght K, et al. Cross-species cellular mapping and humanization of Fcγ receptors to advance antibody modeling. Sci Immunol. 2026;11(115):eady7328. doi:10.1126/sciimmunol.ady7328

[4] Stefanutti E, Ramani R, Whitener B, et al. Analysis of Fc-dependent effector functions of anti-malaria circumsporozoite protein antibodies. Microbiol Spectr. 2025;13(9):e0086325. doi:10.1128/spectrum.00863-25